

in the interior of degenerating cells which have taken up the indicator. The violet change is not due to free carbon dioxide, for bubbling the gas through unclotted plasma containing congo red fails to bring about an alteration in color.

The observations show that tissue cells can withstand much more considerable changes in the reaction of the medium round about than has been supposed; and that their growth *in vitro* may be very active in an acid medium.

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**The location of the optic anlage in *Amblystoma* and the interpretation of certain eye defects.**

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The early embryos of *Amblystoma* were operated upon so as to remove definite areas from the open medullary plate in order to determine the position of the materials which would give origin to the future eyes.

Preliminary experiments of sticking and disturbing the cells in the anterior end of the medullary plate without actually removing these cells did not prevent the subsequent development of the optic vesicles in an apparently normal manner. Cutting out rectangular pieces of the anterior part of the medullary plate, reversing the pieces and transplanting them merely caused the eyes to develop in misplaced positions. These two experiments demonstrate the fact that unless the future eye material is well removed by the operation the optic vesicles may form. This fact is important in considering the results of the following experiments.

A general statement of the results of the experiments may be expressed as follows: Thirty embryos studied after various operations in which lateral portions of the medullary plate were removed at slightly different developmental stages showed in twenty-four individuals, or eighty per cent. of the cases, subsequent development of both eyes, while only six specimens, or twenty per cent.

of the cases, showed absence of the eye. In one case the presence of the eye was questionable, in five cases one eye and in one case both eyes were absent. The absence of the eyes in the latter cases was possibly due to the cut having been made in a more median position than was intended.

Nine embryos studied after having been operated upon so as to remove a narrow median strip of cells from the anterior portion of the medullary plate showed in four cases, or about forty-five per cent. of the specimens, entire absence of eyes. In four other individuals the eyes were highly defective, one specimen having one poorly formed eye while the other was questionably present. In only one of the nine embryos did the eyes approach the normal condition, from this specimen an extremely narrow median piece had been cut out of the medullary plate. The optic anlage might have been sufficiently wide at the time of the operation to allow its median portion to be removed and yet enough material remain on either side of the cut to give origin to the two eyes. According to the views of several investigators the removal of this median material should have caused the cyclopean defect, yet it did not. In a more extended report of these experiments I shall show that cyclopia is not due to a coming together of lateral materials in the median plane, but to a failure of median material to spread laterally.

Contrasting the results obtained after the lateral and median cuts mentioned above one must conclude that: *The eye anlage in the medullary plate occupies an antero-median position as shown by the various abnormalities incurred when this region is cut away. The failure to injure the development of the eyes in the great majority of cases when the lateral portions of the medullary plate are removed by operation indicates further that the eyeanlagen do not occupy lateral positions during this stage of development.*

Based upon these experiments and a study of a large number of eye abnormalities it is concluded that the cyclopean defect is a developmental arrest. The eye anlage fails to widen laterally so that only a single median growth center arises from which develops the ventro-median cyclopean eye. In normal cases the anlage widens and two more or less lateral growth centers become established and give rise to the ventro-lateral optic

vesicles. The optic stalks, however, and later the optic nerves following the stalks as paths always lead back to the point of their median origin and the optic cross or chiasma is in the median plane, below and outside the brain tissue. The attainment of this position of the optic cross would seem mechanically impossible if the eyes arose from lateral medullary tissues since the optic fibers following the stalks would enter the brain laterally and would necessarily cross within the brain tissue, not below and outside as the nerves actually do.

There is no medullary tissue other than future eye tissue between the eye anlagen, therefore, Spemann and others are incorrect in assuming that cyclopia is due to a failure to develop of tissues between the eyes thus permitting the eye anlagen to slump towards the median plane and fuse. The defect is due to a failure or arrest in development of the eye material itself.

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#### **The occurrence of betaine in the muscles of invertebrates.**

**By D. WRIGHT WILSON.**

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Betaine, or trimethylglycocoll, was isolated from the muscle tissue of two varieties of mollusc, *Pecten irradians*, the common scallop, and *Sycotypus canaliculatus*, the periwinkle.

The tissues used were the adductor muscle of the *Pecten* and the large pedal muscle of the *Sycotypus*. The manner of treatment was the same in both cases. The muscles were finely ground, extracted with several changes of water and the concentrated extract freed of colloidal material by precipitation with alcohol and by the regular Kutscher manipulation with tannin. The portion precipitated by phosphotungstic acid was fractionated by precipitation with silver nitrate and barium hydroxide and from the resulting filtrate, betaine was crystallized as the free base and hydrochloride. In both cases, the compound was identified by the melting points of the hydrochloride, picrate and chloroplatinate and by the analyses of the hydrochloride and chloroplatinate.