

can be separated out from the other relatives who, although they manifest abnormal electroencephalographic findings, fail to develop overt seizures. In addition, another important question is whether such cases can be detected in early childhood and preventive measures of some sort taken to eliminate the probability of the onset of overt seizures. Finally, the problem as to the "carrier" nature of such relatives with regard to the heredity of convulsive disorders is a highly important one on which it is hoped the long controlled series of observations now under way at the New York State Psychiatric Institute may shed future light.

Summary. Upon examination of parents and siblings of patients diagnosed as idiopathic epileptics it was shown that 23% of the parents and 28% of the siblings showed definitely abnormal EEG findings. The material fell roughly into 2 groups, that is, those families showing a high incidence of abnormal EEG findings and those families showing no such evidence. The diagnostic, therapeutic and genetic problems are briefly reviewed in relation to these observations.

10850 P

Fluorescence and Absorption Spectra of Flavin Isolated from a Toxic Culture Filtrate of *C. diphtheriae*.

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This note* reports the isolation of flavin elaborated by *C. diphtheriae*, strain No. 3203, grown in synthetic medium. Flavin was separated by ultrafiltration from the toxin and extracted by chloroform from the filtrate after it had been treated with ether to remove the porphyrins soluble in ether and any other ether-soluble substances. The chloroform extract was subjected to chromatographic

* This study is a continuation of previous studies^{1,2,3} of toxigenic growth of the diphtheria bacillus in synthetic medium. I wish to thank Miss Amy Walker for her collaboration in the technical work.

¹ Wadsworth, Augustus, Crowe, M. O'L., and Smith, L. A., *Brit. J. Exp. Path.*, 1935, **16**, 201.

² Wheeler, M. W., and Crowe, M. O'L., *J. Bact.*, 1936, **31**, 519.

³ Crowe, M. O'L., *Proc. Soc. Exp. Biol. and Med.*, 1937, **37**, 215.

analysis and the pigment solutions that were recovered from the column were centrifugalized.

The green fluorescence of the untreated filtrate suggests the presence of flavin. Others^{4, 5} have also noted this fluorescence. The chloroform extract was analyzed on a column of Brockmann's standardized aluminum oxide. When the column was examined in the light of a Hanovia analyzing lamp, several fluorescing zones were observed. The material in the lowest zone, of a brilliant canary yellow fluorescence, was washed from the column with chloroform and, in this solution, fluoresced with a yellow-green light that changed to blue when the solution was exposed to daylight. The material in

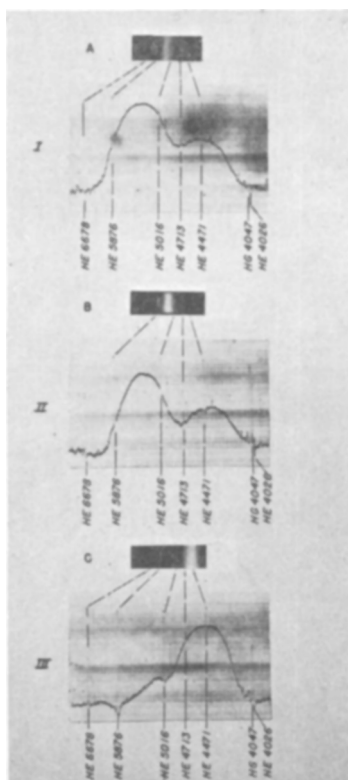


FIG. 1.

Fluorescence spectrograms, A, B, C, of the chloroform extract of an ether-extracted residue of an ultrafiltered diphtheria toxin filtrate; and of the 2 pigments in this extract recovered from a Tswett column in chloroform and methyl alcohol solutions, respectively; and the microphotometric tracings, I, II, and III, of these spectrograms.

⁴ Dhéré, Ch., Meunier, P., and Castelli, V., *C. r. soc. de biol.*, 1938, **127**, 564.

⁵ Dhéré, Ch., and Gourévitch, A., *C. r. soc. de biol.*, 1939, **130**, 593.

the zone above this, which fluoresced with a bright blue light, was elutriated from the column with methyl alcohol and fluoresced in this solution with an intense sky blue light which changed to yellow when the solution was made alkaline and remained so when left standing in daylight. The addition of a reducing agent, sodium hydrosulfite, quenched the fluorescence which was restored when the solution was oxidized by shaking in air.

Uninoculated synthetic medium, when poured on the column, gave a very feeble fluorescence not at all similar to the brilliant yellow and blue light described above.

The fluorescence spectrum of the pigment component of Warburg's yellow enzyme has been reported⁶ as beginning at λ 500m μ and ending indefinitely at λ 630m μ . The fluorescence spectrograms of the chloroform extract and the chloroform and methanol solutions recovered from the column, Fig. 1, A, B, and C, respectively, and their microphotometric tracings, I, II, and III, indicate bands in the same general region between the mercury and helium lines λ 4047 Å and λ 6678 Å, respectively.

While the general contour of the absorption spectrum of the methanol solution recorded on an Eastman panchromatic plate is similar to the curves published for the photoderivative of the pigment component of the yellow enzyme by Warburg and Christian,⁶

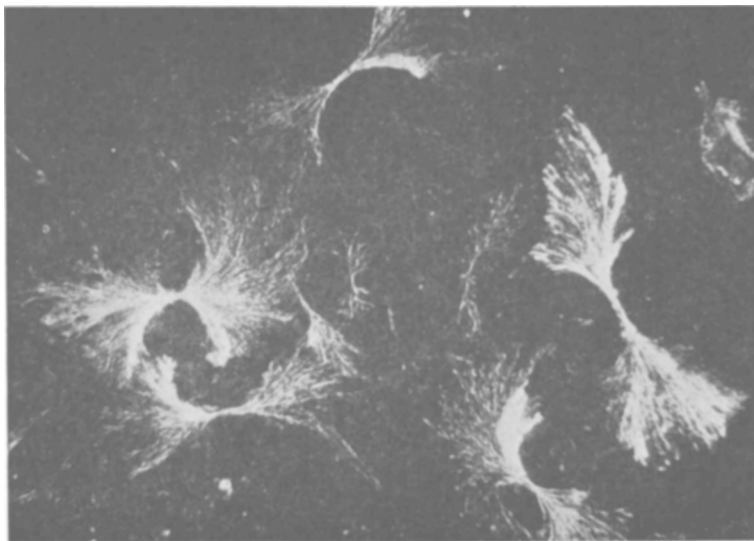


FIG. 2.

Photomicrograph of crystals of the photoderivative of the pigment isolated from diphtheria culture filtrate.

⁶ Warburg, Otto, and Christian, Walter, *Biochem. Z.*, 1933, **266**, 377.

the wave lengths of the positions of selective absorption fall in most instances further towards the violet end of the spectrum at approximately λ 250 $m\mu$, 335 $m\mu$, 370 to 380 $m\mu$, and λ 460 $m\mu$.

The pigment in the concentrations studied is optically inactive or nearly so.†

Crystals were obtained from a water solution of the pigment obtained from the methanol solution. The photomicrograph of the tufts of fine needle-like crystals is presented in Fig. 2 and resembles that of the photoderivative of the pigment component of the yellow enzyme.⁶

Since the tufted arrangement of the crystals in Fig. 2 is somewhat different from that of the photoderivative of the pigment component of Warburg's yellow enzyme, and since the absorption spectrum of the methanol solution of pigment isolated from diphtheria filtrate indicates selective absorption at slightly different wave length values than those published for flavins, it is possible that the flavin synthesized by the diphtheria organism is an unidentified flavin.

10851 P

Experiments on the Mechanism of Gastrulation in a Frog's Egg.

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In the experiments here reported we have attempted to analyze the factors responsible for the turning in (involution) of the dorsal lip of the blastopore. Spemann¹ states that the dorsal lip possesses an inherent capacity to turn in, although under certain abnormal conditions it may fail to do so, growing out into a horn-like projection. Vintemberger^{2, 3} found that the chordal material has a weaker capacity to turn in than the prechordal portion, which lies just below the former. The chordal material is, according to this author, aided in its involution by its attachment to the prechordal material.

† The small quantity of material available for this study did not permit the estimation of the quantity of flavin in the toxic filtrate. Further details will be presented in a report to be published elsewhere.

¹ Spemann, H., *Embryonic Development and Induction*, 1938, p. 105, Yale University Press.

² Vintemberger, P., *Compt. Rend. Soc. de Biol.*, 1938, **127**, 435.

³ *Ibid.*, **127**, 436.