

Making preparations of tissues is greatly facilitated by the use of this medium, and the comparatively constant composition of the mixture renders the results obtained through its use more uniform than those secured by the employment of lymph or plasma. The implanted cells get what very nearly corresponds to their natural food in the serum of the blood, and the gelatine, while apparently acting in no way injuriously to the cells, affords a means of appealing to their thigmotactic proclivities that is ordinarily supplied by the fibrin of clotted plasma.

The outgrowth of epithelium in this medium is remarkable. In some cases it has been over twenty times the superficial area of the implanted tissue. As a rule the tissues thrive better than in plasma or lymph. It is comparatively easy to subculture the tissues, since the gelatine dissolves in Ringer's solution, and by washing the preparations in this fluid they may be readily freed, and then transferred to a fresh culture medium. I have transferred pieces of epithelial tissue several times in succession, and kept them thriving for three months. Cell divisions have been repeatedly seen in epithelial cells in this medium. In a piece of tissue put up on February 17 and changed to fresh culture fluid three times afterwards, I observed several mitotic figures in epithelial cells on April 8, fifty days after the preparation was made. The chromosomes could be seen with great distinctness in the living material. In one cell first seen in the prophases of division, the chromosomes were seen to align themselves in the equatorial plate, then to be drawn apart, and finally to become constituted into the two daughter nuclei; at the same time the constriction in two of the cell body could be distinctly followed. Over a dozen other mitotic figures in various stages were observed in the same piece. The preparation had been washed in Ringer's solution and transferred to new culture medium a few days previously, after which it had taken on a new lease of life. The division figures were all seen in a transparent sheet of epithelium that had spread out in contact with

the cover slip supporting the hanging drop culture.

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#### ON THE CHEMICAL NATURE OF THE LUMINOUS MATERIAL OF THE FIREFLY

OUR knowledge of the chemistry of light production by organisms may be summed up in the statement that phosphorescence is due to the oxidation of some substance formed in the cells of the animal. As with other oxidations, both water and oxygen must be present. If either water or oxygen are absent the photogenic substance will not be used up by oxidation. Luminous tissues if dried rapidly may be ground up and preserved indefinitely, and at any later time, if moistened in the presence of oxygen, will phosphoresce. This old and important discovery makes the investigation of the chemical nature of the luminous substance relatively easy. The dried powder of the luminous organ may be extracted with: (1) Oxygen-free watery solvents, or (2) water-free solvents (as ether, chloroform, etc.) with or without oxygen.

The earlier workers supposed the photogenic material to be phosphorus or phosphine. These views require no comment to-day. Later suggestions have been that the substance is a fat, an albumin, a lipid (lecithin), a nucleoalbumin or a lecithoprotein (phosphatid). It is obviously desirable to know whether the substance is fat-like or protein in nature. The fact that phosphorescence ceases as soon as the moist luminous material is heated to 100° proves nothing, for, like organic oxidation in general, an oxidizing ferment is probably involved, and it is this oxidase which may be destroyed on heating.

I can state definitely that the "luciferin" of the common fire-fly is not a true fat or any fat-like body such as lecithin. The dried material may be extracted with anhydrous ether and the ether extract evaporated to dryness. On adding water or a watery extract of luminous organ (to add an oxidizing enzyme) or potato juice (to add an oxidase) to the residue no phosphorescence took place; on adding water to the original ether extracted

material brilliant phosphorescence occurred. The same results were obtained with anhydrous chloroform, ethyl alcohol, acetone and carbon tetrachloride. The material is therefore insoluble in fat solvents.

It is most likely a protein but belongs among the proteins insoluble in water. By means of a specially constructed apparatus I was able to extract with oxygen-free distilled water and to filter the extract in an oxygen-free space. On admitting air the filtrate did not glow, but the filter paper showed innumerable bright dots. The granules of luminous substance are therefore insoluble in water. A lack of material has prevented extraction with other protein solvents, salt solution, acids and alkalies.

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#### THE AMERICAN CHEMICAL SOCIETY. II

##### DIVISION OF FERTILIZER CHEMISTRY

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Chairman's Address: *Chemistry an Important Factor in the Fertilizer Industry*: J. E. BRECKENRIDGE.

*The Preparation of Neutral Ammonium Citrate*: ERMON D. EASTMAN AND JOEL H. HILDEBRAND.

The proposed method depends on the preparation of a standard sodium phosphate solution of known hydrogen ion concentration and the comparison of the color produced by rosolic acid in this solution with that produced by the same indicator in the ammonium citrate solution to be tested. The normal ammonium citrate solution is shown by its hydrogen ion concentration to be slightly acid and the authors have therefore adopted the neutral rather than the normal solution.

*A Comparison of Neutral Ammonium Citrate with Sodium Citrate and N/10 Citric Acid*: PAUL RUDNICK, W. B. DERBY AND W. L. LATSHAW.

Sodium citrate proposed by Bosworth (2) can be used as a substitute for the official neutral ammonium citrate, but N/10 citric acid obviates difficulties due to highly concentrated solutions, such as slowness in filtration, etc., and gives results which are in excellent agreement with those obtained by the official neutral ammonium citrate.

*The Separation of Organic Nitrogen from Mixed Fertilizers*: C. H. JONES.

The method recommended depends on separation by gravity in carbon tetrachloride. Tables giving the behavior of various fertilizer ingredients and their availability by the alkaline permanganate method are included.

*Separation of Phosphoric Acid from Lime*: F. K. CAMERON.

A discussion of the solubility curves of potassium and ammonium phosphates and their applications to practical problems.

*Separation of Potash from Kelp (lantern)*: F. K. CAMERON.

An illustrated description of the kelp beds and the methods of harvesting so far developed.

##### DIVISION OF PHARMACEUTICAL CHEMISTRY

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*Methods of Analysis of the Forthcoming Pharmacopœia*: H. W. WILEY.

*Seasonal Variation in the Composition of the Thyroid Gland*: ATHERTON SEIDELL AND FREDERIC FENGER.

The experiments upon this subject embracing the period August, 1911, to August, 1912, have been continued for another one-year period beginning December 1, 1912. The evidence for the seasonal variation in iodine content of the thyroid gland has been confirmed, and additional data obtained, showing that a regular change of phosphorus and ash, varying inversely with the iodine, occurs. In regard to the fresh weight of the glands, the results indicated a regular seasonal change in the case of the beef and sheep, but not with the hog. The results demonstrate the practicability of a standard of 0.2 per cent. iodine in commercial desiccated thyroids.

*Some Peculiarities of Present Food and Drug Laws*: FRANK O. TAYLOR.

*Notes on the Determination of Antipyrine*: GEORGE D. BEAL AND DUANE T. ENGLIS.

Antipyrine and caffeine can be easily extracted by chloroform from an aqueous solution three-fourths saturated with sodium chloride. If the liquid contains vegetable extractives, the extraction can be effected without emulsification by first precipitating the coloring matter, resins, etc., with lead acetate. The antipyrine may be titrated in the presence of caffeine by Bougault's<sup>1</sup> method,

<sup>1</sup> *Jour. Pharm. Chem.*, [6], 1, 161, 11, 97.