The addition of some beeswax and resin to the paraffin-lampblack mixture will increase to a considerable extent the rigidity of the plate, although these constituents are not absolutely necessary.

Before the plate has hardened, and is still plastic, the anatomical specimen is pressed down slightly into the soft plate to hold it in position and is fastened there with thread looped around the parts of the specimen, passed through holes, and tied at the back of the plate. Necessary labels are now attached.

When completed, the preparation is immersed in a preservative in a museum jar. Such preparations will last for an indefinite time.

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SPECIAL ARTICLES

THE PARTICLE SIZE OF THE VIRUS OF EQUINE ENCEPHALOMYELITIS

A MOST interesting epidemic of acute encephalomyelitis occurred among horses and mules in various local districts of California during the summer and fall of 1930, 1931 and 1932. Bacteriological and pathological studies of the disease were conducted by Meyer, Haring and Howitt and these workers have reported elsewhere[1,2,3] the salient characteristics of the virus and its effects on animals. In this work they recorded the filterability of the virus through Berkefeld V and N candles. However, filtration experiments utilizing the usual forms of candles, furnish no adequate basis for estimating the particle size of the virus because many factors other than mechanical sieve action have been shown to condition the filterability of a microorganism through such filters.

In order to obtain some idea of the virus particle size filtration experiments were performed with the acetic collodion gel ultrafilter series described by Krueger and Ritter[4] which possess the advantage of uniformity and low adsorbing surface area. The pore sizes of these membranes depends upon the percentage of nitro-cellulose dissolved in the glacial acetic acid and have been estimated by testing the permeability both to colloidal sols of known particle size and to water under certain standard conditions. The two methods of estimation give figures of the same order of magnitude with a relatively small constant difference between them. Manegold and Veit[5] have shown that this difference may be materially reduced by basing the pore size calculations from water permeability data upon the assumption of a random pore distribution as contrasted to parallel capillary bundles such as we had assumed to exist in the membranes. The result is a practical coincidence of the two pore size curves.

During the past year and one-half we have used different materials in making the membranes and have made minor alternations in the technique of preparation. The pore sizes of the series necessarily

ULTRAFILTRATION OF SUSPENSIONS OF ENCEPHALOMYELITIS BRAINS

No.	Dilution of original supernatant from 20 per cent. brain suspension	Per cent. collodion (Ultrafilter)	Bacterial cultures from filtrate	Animal inoculation filtrate
1	Undiluted	3.0	0	Typical disease
2	Undiluted	3.0	0	Typical disease
3	1:2 with saline	3.0	B. subtilis	Typical disease
4	1:2 with saline	3.0	B. subtilis	Negative
5	Control undiluted	Unfiltered	0	Typical disease
6	Control 1:2 with saline	Unfiltered	0	Typical disease
7	1:2 with broth	3.0	0	Typical disease
8	1:2 with broth	3.0	0	Typical disease
9	Control undiluted	Unfiltered	0	Typical disease
10	Control 1:2 with broth	Unfiltered	0	Typical disease
11	Undiluted	4.0	0	Negative
12	Undiluted	4.0	0	Negative
13	Control, undiluted	Unfiltered	0	Typical disease
14	Control, undiluted	Unfiltered	0	Typical disease
15	Undiluted	3.5	0	Negative
16	Undiluted	3.5	0	Negative
17	Control, undiluted	Unfiltered	0	Typical disease
18	Control, undiluted	Unfiltered	0	Typical disease
19	1:2 with broth	3.5	0	Negative
20	1:2 with broth	3.5	0	Negative
21	Control, undiluted	Unfiltered	0	Typical disease
22	Control 1:2 with broth	Unfiltered	0	Typical disease

[1] K. F. Meyer, C. M. Haring and B. Howitt, "The Etiology of Epizootic Encephalomyelitis of Horses in the San Joaquin Valley, 1930," SCIENCE, 74: 227–228, 1931.

[2] C. M. Haring, J. A. Howarth and K. F. Meyer, "An Infectious Brain Disease of Horses and Mules." (Encephalomyelitis)," University of California Agricultural Experiment Station Circular 322, August, 1931.

[3] B. Howitt, "Cross Immunization Experiments with Poliomyelitis Virus and that of Encephalomyelitis in Horses," *Proc.* Soc. Exp. Biol. and Med., 29: 118–120, 1931.

[4] A. P. Krueger and Ritter, "The Preparation of a Graded Series of Ultrafilters and Measurement of their Pore Size," *Jour. Gen. Physiol.*, 13; 409, 1930.

[5] E. Manegold and K. Veit, "Über Kapillarsysteme, XI," Kol. Zeitschr., Bd. 56, H. 1, 1931.

have had to be re-determined and do not correspond to the previously reported series. A description of the present membranes will be reported elsewhere.

In the present experiments the test material used was a 20 per cent brain suspension in normal saline freshly prepared from guinea-pigs which had been killed at the height of the disease. The suspension was centrifuged at low speed to remove coarse tissue particles and was then filtered under low negative pressures through sterile membranes of various pore sizes. The filtrates were tested for the presence of the virus by inoculating 0.3 cc. intracerebrally into guinea-pigs.

The table appended shows that the virus traverses a 3 per cent. membrane but is retained by a 3.5 per cent. membrane. The usual bacteriological tests for the detection of microorganisms were applied to the filtrates with negative results. Our data would indicate an approximate particle size of 500 μμ for the virus as it exists in brain suspensions. Under like conditions of preparation and filtration it is of the same order of magnitude as the causal agent of poliomyelitis[6] an analagous disease of man and is apparently ten times the size of the hoof and mouth disease virus particle.[7] Dilution does not appear to render the virus more finely dispersed, as by elution from carrier particles for example, nor to affect the filter pore surfaces so that the particles pass more readily.

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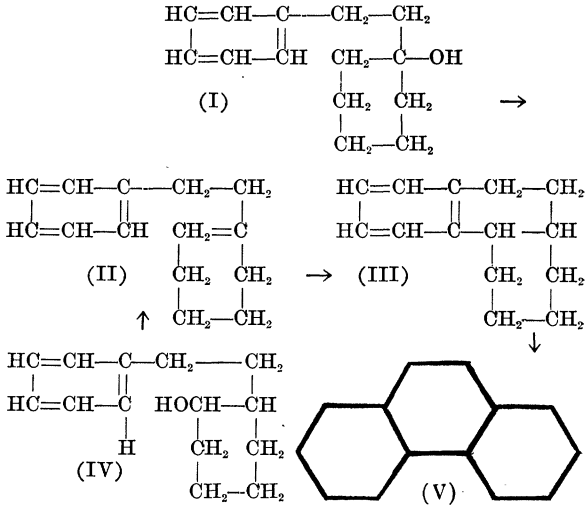
A NEW PROCESS FOR THE SYNTHESIS OF PHENANTHRENE AND OF PHENANTHRENE DERIVATIVES

FOLLOWING up the researches on ionene lately communicated,[1] Davidson and Perlman have now succeeded, in these laboratories, in applying the knowledge gained in the ionene field to the development of a new process for the synthesis of phenanthrene and of its derivatives.

By the well-known Grignard reaction, 1-phenethyl-cyclohexanol-1 (I) was prepared from phenethyl magnesium bromide and cyclohexanone. When this tertiary alcohol (I) was treated with concentrated sulfuric acid, it split out a molecule of water and condensed to the same octahydrophenanthrene (III) as Bardhan and Sengupta[2] obtained from 1-phenethyl-cyclohexanol-2 (IV), and like it gave a good yield of phenanthrene (V) when dehydrogenated by heating with selenium.

Bardhan and Sengupta appear to assume a direct cyclodehydration between the OH of the alcohol and an H of the benzene nucleus. But, as we have shown in the ionene group (see the above reference), such condensations may proceed rather through the formation of the olefin first, which then rearranges by cyclization. That this is the case here also, Davidson and Perlman have now proven by heating the alcohol (I) for one minute with 50 per cent. sulfuric acid, when the olefin (II) was obtained. The same olefin was prepared by distilling the alcohol with a crystal of iodine. Treatment of this olefin with concentrated sulfuric acid rearranged it to the octahydrophenanthrene (III). These reactions indicate that the structure of the olefin is that shown in the formula (II), and that such an olefin could be formed by dehydration of either our alcohol (I) or that of Bardhan and Sengupta (IV). If this is really the mechanism of the condensation, as we believe it to be, it is clear why the same octahydrophenanthrene results whether the OH on the cyclohexane nucleus is in Position 1 (I) or 2 (IV).



We believe that this synthesis has certain advantages over that of Bardhan and Sengupta, in that it starts with a simpler initial material, cyclohexanone instead of the potassium derivative of its ethyl carboxylate, and involves fewer steps. The investigation therefore is being extended in various directions, by using other carbonyl compounds in place of cyclohexanone and and other Grignard reagents instead of phenethyl magnesium halides.

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[6] A. P. Krueger and E. W. Schultz, "Ultrafiltration Studies on the Virus of Poliomyelitis," *Proc.* Soc. Exp. Biol. and Med., 26: 600, 1929.

[7] P. K. Olitsky and L. Boez, *Jour. Exp. Med.*, 45: 673, 1927.

[1] Bogert, SCIENCE, n. s., 76: 1977, 475, November 18, 1932.

[2] *Jour. Chem. Soc.*, 1932, 2520.