have begun a study of methods of combining entrance requirements in its three branches to go into effect next autumn. The Institute of Technology is composed of the College of Engineering and Architecture, the School of Chemistry and the School of Mines and Metallurgy. M. Cannon Sneed, chief of the division of inorganic chemistry, is chairman of the committee on registration, entrance requirements and curriculum for first-year students. Other members of the committee are Professor Elting H. Comstock, of the mines faculty; Robert W. French and I. W. Geiger, associate professors of drawing and chemistry, respectively, and Charles A. Koepke, associate professor of mechanical engineering. Professor Geiger; W. E. Brooke, head of the department of mathematics and mechanics; Howard D. Myers, associate professor of engineering; Professor W. T. Ryan, electrical engineering, and Professor Comstock will study entrance requirements. Chairman of the group studying a common curriculum for all first-year students in the institute is Professor W. H. Kirchner, head of the department of drawing and descriptive geometry. Others are Professor Comstock; Henry C. Eggers, assistant professor of drawing; F. M. Mann, head of the School of Architecture, R. E. Montonna, associate professor of of chemical engineering; Professor John R. DuPriest, head of the department of mechanical engineering, and Dean Ora M. Leland, of the College of Engineering and Architecture.

Nature states that two British expeditions to observe the total eclipse of the sun on June 19 are leaving for sites selected from which to observe the eclipse. The path of the total eclipse stretches from Greece over Siberia to the Pacific Ocean. An expedition led by Professor F. J. M. Stratton, of the Solar Physics Observatory, Cambridge, will station itself in northern Japan. The program of eclipse observations consists chiefly of observations of intensities of lines in the flash spectrum; despite the vigorous growth of the technique of spectrophotometry in the last decade, very few spectrophotometric observations have been made on eclipses, chiefly on account of the ill-luck through cloud which has attended recent expeditions. The second British expedition will be led by Professor J. A. Carroll, of the University of Aberdeen, and will proceed to a site in the U.S.S.R. where the eclipse will take place near midday.

## DISCUSSION

## OBSERVATIONS ON THE CULTIVATION OF POLIOMYELITIS VIRUS

IT is stated generally and is to some extent accepted as a fact that filterable viruses can not be cultivated on ordinary lifeless media. While many of these ultramicroscopic forms have been observed to multiply in vitro in the presence of living susceptible cells or in modified tissue culture media, these successes seem to build all the more solidly on the postulate that the viruses are probably obligate parasites. The characteristics, distinguishing peculiarities and the techniques required for the demonstration and study of viruses and their behavior have been described fully by Rivers<sup>1</sup> and collaborators in this country and by Bedson<sup>2</sup> and associates in England. On the basis of their findings one is compelled to accept as a dictumno virus multiplication without the presence of viable or susceptible cells or tissue in a culture medium.

Of particular interest in this connection are the

<sup>1</sup>T. M. Rivers, The Harvey Lectures, 1933-34, Baltimore, Williams and Wilkins Company; *Pennsylvania Med. Jour.*, April, 1933; *Am. Jour. Med. Sci.*, 190: 435, 1935; Rivers' Filterable Viruses, 1928, Baltimore, Williams and Wilkins Company; *Jour. Exp. Med.*, 52-60, 1930-35.

<sup>2</sup>S. P. Bedson, Brit. Jour. Exp. Path., 8: 470, 1927; 11: 502, 1930; 13: 65, 1932; 14: 267, 1933; 15: 243, 1934; Newcastle Med. Jour., 15: 55; 105, 1935; The Lancet, 1277, December 7, 1935. reports of a few investigators who brought forth evidence which was at variance with the definition of a virus, notably regarding its requirements for actual multiplication outside the animal body. Eagles and McClean,<sup>8</sup> working with vaccine virus, claimed to have cultivated the virus in a "cell-free" medium containing extract of rabbit kidney tissue, serum and Tyrode's solution. Rivers and Ward<sup>4</sup> were unable to confirm these results and succeeded shortly afterward in establishing the correctness of the observations of Maitland<sup>5</sup> and coworkers, who had previously shown that it was possible to accomplish this only in the presence of minced kidney tissue. It followed from subsequent studies by Rivers<sup>6</sup> that splenic and testicular tissue could also supply the necessary elements for growth of vaccine virus. Eberson,<sup>7</sup> in an attempt to cultivate the virus of poliomyelitis in a medium containing

<sup>8</sup> G. H. Eagles and D. McClean, Brit. Jour. Exp. Path., 11: 337, 1930; 12: 97, 1931; see also G. H. Eagles and A. H. H. Kordi, Proc. Roy. Soc. London, Series B, 111: 329, 1932.

<sup>4</sup>T. M. Rivers and S. M. Ward, *Jour. Exp. Med.*, 57: 51, 1933.

<sup>5</sup> H. B. Maitland and M. C. Maitland, *Lancet*, 2: 596, 1928; *Brit. Jour. Exp. Path.*, 11: 119, 1930; 13: 90, 1932. <sup>6</sup> T. M. Rivers and S. M. Ward, *Jour. Exp. Med.*, 57: 741, 1933.

τ<sup>'</sup>F. Eberson, Proc. Soc. Exp. Biol. and Med., 29: 477, 1932; SCIENCE, 75: 519, 1932; Proc. Soc. Exp. Biol. and Med., 30: 92, 1932; Jour. Lab. and Clin. Med., 18: 565, 1933; Jour. Immunol., 24: 433, 1933.

macerated sheep brain, successfully transmitted the disease in serial passages through Macacus rhesus monkeys by means of cultures of the virus in dilutions ranging from  $2 \times 10^{-7}$  to  $2 \times 10^{-27}$  of the original material.

In view of what has been discussed it is now apparent that the asserted lifeless culture medium of Eberson can not be regarded as such. The observations and conclusions drawn from that study are explainable only on the basis of a modified tissue culture which was capable of supporting the existence of poliomyelitis virus and enabling it to multiply in the presence of viable and susceptible cells. Hence the results which embodied transmissibility in series, infectivity and immunological considerations were not surprising or unexpected. A reexamination of the original protocols and a scrutiny of the photomicrographs which illustrated the article indicate clearly that the visible bodies or "organisms" multiplied in and about the tissue particles, the nuclear elements especially. This was stated unequivocally in the text with regard not only to the subculturing of the material in serial dilutions, but also with reference to the type of inoculum employed in the transmission of the disease to monkeys. An experiment performed in 1931 at the Rockefeller Institute,<sup>8</sup> with a submitted culture in the eighth subplant representing a dilution of the original inoculum of approximately  $2 \times 10^{-17}$ , emphasized this point. There it was decided to inject intracerebrally in a monkey one cubic centimeter of the supernatant fluid from lightly centrifuged culture material which had previously been ground in a mortar with sterile quartz sand. The infective power of such a culture was successfully demonstrated when poliomyelitis developed after one week in the test animal, from which in turn it was possible to transmit the infection to another monkey with material derived from a suspension of brain and nervous tissue. The intimate relation of the multiplying virus to the tissue particles of the culture medium was thus indubitably shown.

With regard to the brain tissue medium itself it was stated that it must have been lifeless in consequence of the mode of sterilization. This assumption, despite subsequent thermal controls, was erroneous. Adequate heat penetration was made difficult, owing to the nature of the medium and its containers during the process of sterilization. That there was considerable variation in the different lots of medium as a result of this would follow from the irregular successes of some infectivity experiments, particularly in the later subplants from cultures beyond the tenth generation. Supplementary experiments designed to cultivate the virus with thoroughly cooked culture medium prepared in another laboratory resulted in failure. This

<sup>8</sup> F. Eberson, Jour. Lab. and Clin. Med., 18: 586, 1933.

was to be expected from the nature of the virus and thus confirmed the fact that the substrate used originally could not have been lifeless in the accepted sense.

Regardless of present or future attempts to cultivate certain viruses in lifeless media, and assuming this as only remotely possible, much knowledge concerning their behavior and properties can be gained nevertheless by a study of viruses in a tissue culture medium or some modification of it. This has been amply demonstrated recently for the infective agents of vesicular stomatitis,<sup>9</sup> poliomvelitis,<sup>10</sup> psittacosis<sup>11</sup> and louping ill,<sup>12</sup> all these having yielded to cultivation in media similar to that used for vaccine virus.

In discussing this subject it is recognized that the difficulty in cultivating a virus is related directly to its degree of parasitism. Subject to this condition it should be possible to devise methods of study with culture media adapted to the needs of individual viruses. Considering their widely divergent behavior in the animal and human tissues, is it not paradoxical to suppose that all viruses in common should conform to a single type of cell-host parasitism?

A study is now in progress to determine whether or not the virus of poliomyelitis can be adapted to cultivation in various modifications of culture media containing suitable tissue and physiological fluids. It should be of some interest to ascertain the possible relationship between the ability of a given virus to multiply and its reputed degree of parasitism, to the end that a "parasitic index" for viruses in general might be evaluated.

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## THE VITAMIN C CONTENT OF APPLES AND ITS RELATION TO HUMAN WELFARE

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In view of the extreme variety differences in vitamin C content of apples<sup>1, 2, 8, 4</sup> it has been pointed out<sup>5</sup> that the exchange of certain varieties of apples

9 H. R. Cox, J. T. Syverton and P. K. Olitsky, Proc. Soc. Exp. Biol. and Med., 30: 896, 1933.

<sup>10</sup> E. Gildemeister, Cent. Bakt., Abt., 1, Ref., 109: 284, 1933.

<sup>11</sup> S. P. Bedson and J. O. W. Bland, Brit. Jour. Exp. Path., 15: 246, 1934; J. O. W. Bland and R. G. Canti, Jour. Path. and Bact., 40: 231, 1935.

<sup>12</sup> T. M. Rivers and S. M. Ward, Proc. Soc. Exp. Biol. and Med., 30: 1300, 1933.

1 M. F. Bracewell, E. Hoyle and S. S. Zilva, Biochem. Jour., 24: 82-90, 1930.

<sup>2</sup> M. F. Bracewell, E. Hoyle and S. S. Zilva, British

Med. Res. Council, Sp. Rpt. Ser. B., 146: 3-145, 1930. <sup>3</sup> C. R. Fellers, P. D. Isham and G. G. Smith, Proc. Am. Soc. Hort. Sci., 29: 93-97, 1932.

4 G. G. Smith and C. R. Fellers, Proc. Am. Soc. Hort. Sci., 31: 89-95, 1934.

<sup>5</sup>W. Franklin Dove, Am. Nat., 69: 469-544, p. 524, 1935.