

suffered no modification, has now, through repeated stretching, been so greatly modified that it has lost its identity as a surface covering (Fig. 1e). In order to escape the rather incongruous situation of switching from a fiber which is initially elastic because of the protein composition of its surface, to a later stage in the transformation of that fiber which is now elastic because of a protein-hydrocarbon complex, it is only necessary to assume that hydrocarbon enters into the composition of the original membrane. The feeble zeta potential on Cryptostegia latex globules, and the dissimilarity between the migration curves of latex globules and serum proteins, give experimental support to this view.

The foregoing deduction is in keeping with much work on the chemical nature of the surfaces of living

cells and natural emulsions. Biologists have long regarded cells as coated with fatty substances. Recently, the milk globule, heretofore thought to be stabilized by casein, has been shown by Palmer<sup>2</sup> and by Moyer<sup>3</sup> to be a complex of phospholipids and protein. Moyer's work on milk is very similar, both as to method and the conclusion reached, to the study on latex reported here.

It is difficult to escape the conclusion that molecular continuity exists between the core and the surface of latex globules, and that the elasticity of crude rubber when first formed resides in a surface layer which is a composite of protein and hydrocarbon.

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## SCIENTIFIC APPARATUS AND LABORATORY METHODS

### SEDIMENTATION OF POLIOMYELITIS VIRUS BY CENTRIFUGATION<sup>1</sup>

NUMEROUS reports have appeared in the literature showing that the virus of poliomyelitis can be sedimented by ultracentrifugation. The original report of Schultz and Raffel,<sup>2</sup> who effected partial purification from infected monkey spinal cord, has been amply confirmed, both for human virus and for the spontaneous mouse poliomyelitis of Theiler.<sup>3,4,5,6</sup> The method has been adapted for the recovery of virus from stools by Melnick,<sup>7</sup> and more recently the same author has demonstrated the presence of virus in the blood of experimentally infected monkeys by this technic.<sup>8</sup>

In all these reports, air-driven ultracentrifuges have been used, running at speeds of 22,000 to 39,000 r.p.m. for periods of one to six hours. It occurred to us that prolonged centrifugation at lower speeds, in an apparatus more readily obtainable and less expensive, might be effective in sedimenting virus.

For this purpose, experiments have been performed using the International Equipment Company Multi-speed Attachment on an ordinary Size 2 centrifuge. The head of this attachment, six inches in diameter, holds six tubes at a 45° angle, with a total capacity

of 33 cc. Speeds up to 18,000 r.p.m. are obtainable, at which a force 25,000 times gravity is exerted.

#### EXPERIMENTAL

For all experiments the mouse-adapted Lansing strain of poliomyelitis virus was used. A 20 per cent. stock suspension was prepared in saline and centrifuged at 18,000 r.p.m. for 20 minutes, the clear supernate being stored in ampoules on dry ice. On thawing, some particulate matter appeared which was removed by re-centrifugation at 18,000 r.p.m. for 10 minutes before the experimental tests. For the latter, a period of four hours at 18,000 r.p.m. was arbitrarily selected. Shorter periods were not employed. To keep the centrifuge head below room temperature, the centrifuge chamber was cooled by the addition of about five pounds of dry ice. Centrifugation was interrupted hourly to replenish the supply. Experiments were performed with 20 per cent. and with 1 per cent. suspensions, both in saline.

With 20 per cent. virus a small, transparent, amber-colored pellet was formed. With 1 per cent. virus, a film was deposited over the lower surface of the tube. After centrifugation, the tubes were washed twice with saline, without disturbing the sediment. The gummy sediment was then triturated with a glass rod in a drop of saline, and diluted with saline to the original volume. Resolution was not complete, and in the case of 20 per cent. virus the reconstituted sediment was centrifuged at low speed for a few minutes before diluting it for titration, a certain amount of virus presumably being lost. Mice were inoculated intracerebrally with 0.03 cc of the clear supernate.

The results of four experiments are summarized in Table 1. In the first, the top and bottom thirds of

<sup>1</sup> Aided by a grant from The National Foundation for Infantile Paralysis, Inc.

<sup>2</sup> E. W. Schultz and S. Raffel, *Proc. Soc. Exp. Biol. and Med.*, 37: 297, 1937.

<sup>3</sup> P. F. Clark, A. F. Rasmussen, Jr., and W. C. White, *Jour. Bact.*, 42: 63, 1941.

<sup>4</sup> H. S. Loring and C. E. Schwerdt, *Jour. Exp. Med.*, 75: 395, 1942.

<sup>5</sup> S. Gard and K. O. Pederson, *SCIENCE*, 94: 493, 1941.

<sup>6</sup> J. L. Melnick, *Proc. Soc. Exp. Biol. and Med.*, 49: 553, 1942.

<sup>7</sup> J. L. Melnick, *Jour. Exp. Med.*, 77: 195, 1943.

<sup>8</sup> J. L. Melnick, *Proc. Soc. Exp. Biol. and Med.*, 58: 14, 1945.

<sup>2</sup> *Jour. Biol. Chem.*, 104: 359, 1934.

<sup>3</sup> *Jour. Biol. Chem.*, 133: 29, 1940.

TABLE 1  
RESULTS OF CENTRIFUGATION OF 20 PER CENT. AND 1 PER CENT. VIRUS SUSPENSIONS IN SALINE

| Experiment number | Concentration centrifuged Per cent. | Concentrations of original tissue |                  |                  |                  |                  |                   |                  |                  |                  |                  |                  |
|-------------------|-------------------------------------|-----------------------------------|------------------|------------------|------------------|------------------|-------------------|------------------|------------------|------------------|------------------|------------------|
|                   |                                     | Control virus                     |                  |                  |                  | Supernate        |                   |                  |                  | Sediment         |                  |                  |
|                   |                                     | 10 <sup>-1</sup>                  | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-1</sup> | 10 <sup>-2</sup>  | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> |
| I                 | { 1—Top third<br>1—Bottom third     |                                   |                  |                  |                  |                  | 8/8<br>2/8<br>4/8 | 2/8<br>1/8       |                  | 6/8              | 5/8              | 0/8              |
| II                | 20                                  | 5/6                               | 4/6              | 2/5              |                  | 2/6              | 0/6               |                  |                  | 6/6              | 3/5              | 3/6              |
| III               | { 20<br>1                           |                                   | 5/6              | 3/6              | 2/6              | 0/6<br>2/6       | 0/6<br>2/6        | 1/6<br>0/6       | 0/6<br>2/6       | 6/6<br>2/6       | 0/6<br>1/6       | 2/6<br>0/6       |
| IV                | { 20<br>1                           |                                   | 7/9              | 5/10             | 1/10             | 1/10<br>5/10     | 1/10<br>5/10      | 0/10<br>1/10     | 0/10<br>4/10     | 9/10<br>4/10     | 5/10<br>1/10     | 0/10<br>0/10     |

Numerator: number of mice with proven poliomyelitis.  
Denominator: number of mice injected.

the supernate were tested separately, to determine whether concentration rather than sedimentation might have taken place. Supernate after the preliminary centrifugation was used as control virus in each experiment. In the table, all dilutions refer to concentration of original tissue.

With only a few exceptions, all mice found dead without previously observed symptoms, during a period of five weeks after inoculation, were studied histologically. Sections of brain and cord were examined at 0.8 mm intervals. Only those with paralysis or with typical lesions were considered as having had poliomyelitis.

It will be seen from Table 1 that when 20 per cent. virus was centrifuged, very little activity remained in the supernate, and there was almost quantitative recovery in the sediment. With 1 per cent. virus, considerable amounts remained in the supernate, although virus could be recovered in the sediment.

The effect of homologous normal serum, one-tenth volume, in promoting sedimentation was next tested. The comparative effects of saline and of serum, respectively, on sedimentation, using 1 per cent. virus, are shown in Table 2. The presence of serum results in the formation of a discrete pellet, similar to that formed from more concentrated virus suspensions. The results of this experiment indicate that virus can be recovered quantitatively in the sediment from dilute suspension by the addition of serum.

The purpose of the study was to develop a simplified technic for recovery of virus from biological material. Four stools from human paralytic poliomyelitis patients, in the first week of the disease, have been tested. Heavy suspensions of stool were prepared in water. After preliminary centrifugation at low speed and at 18,000 r.p.m. for 30 minutes, the supernate was centrifuged at 18,000 r.p.m. for four hours. A small translucent or opaque pellet formed. It was treated as described and diluted to a total volume of 1.8 cc in saline. The resulting suspension was treated with ether for from 4 to 18 hours. Two rhesus monkeys for each stool were inoculated with

0.8 cc each by the direct intrathalamic route of Howe and Bodian.<sup>9</sup> No toxic effects were encountered. As controls, two rhesus monkeys for each stool were given daily intranasal instillations of untreated suspension for 4 to 10 days.

Two specimens were positive by both methods, two

TABLE 2  
COMPARATIVE RESULTS OF CENTRIFUGATION OF 1 PER CENT. VIRUS SUSPENSIONS IN SALINE AND IN 10 PER CENT. NORMAL SERUM

|                           |                         | Dilutions of original tissue |                  |                  |
|---------------------------|-------------------------|------------------------------|------------------|------------------|
|                           |                         | 10 <sup>-2</sup>             | 10 <sup>-3</sup> | 10 <sup>-4</sup> |
| Control virus (in saline) |                         | 5/10                         | 3/10             | 1/10             |
| Centrifuged in saline     | { Supernate<br>Sediment | 7/10<br>9/10                 | 5/10<br>2/10     | 1/10<br>0/10     |
| Centrifuged in serum      | { Supernate<br>Sediment | 0/10<br>10/10                | 0/10<br>6/10     | 0/10<br>3/10     |

Numerator: number of mice with proven poliomyelitis.  
Denominator: number of mice injected.

negative by both. Thus, the results are inconclusive so far as the use of this technic is concerned for the detection of virus in biological material where it might otherwise be missed. It does, however, offer a simplified means for the preparation of material for intracerebral inoculation, for which bacteria and toxic substances must be removed. No experiments have been performed in which stool suspensions were centrifuged in the presence of normal serum.

#### SUMMARY

A technic is described whereby poliomyelitis virus may be sedimented by centrifugation for four hours at 18,000 r.p.m.

Virus has been recovered quantitatively in the sediment from a 20 per cent., but not a 1 per cent., saline suspension. The addition of 10 per cent. normal serum results in quantitative recovery from a 1 per cent. suspension.

Virus has been recovered by intracerebral inocula-

<sup>9</sup> H. A. Howe, H. A. Wenner, D. Bodian and K. Maxey, *Proc. Soc. Exp. Biol. and Med.*, 56: 171, 1944.

tion from two of four human stools tested by the method described.

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### NEW PROTECTION FOR FIELD DATA

MAPPING of vegetational types on the U. S. Fish and Wildlife Service's Patuxent Research Refuge, Bowie, Maryland, led to a search for a protective covering for field maps. The problem was solved by a commercially available acetate sheeting treated with a pressure sensitive adhesive. Developed as an easily applied protection for maps for the Army and Navy and commended after ample trial overseas, this material promises to be a boon to scientists who have long suffered with muddy maps and dog-eared data sheets. Easily peeled from the backing attached in manufacture, the film is simply smoothed with thumb-nail pressure onto aerial photos, topographic or vegetational maps, relative humidity tables and other data subjected to repeated use, furnishing a transparent and waterproof covering.

While two types are available, clear and matte, the matte variety is quite transparent enough for most purposes and has the great advantage that it can be marked with pencil, crayon or ink (in washes as well as lines). Such marks adhere well but can be easily erased without damaging the surface. In this way, numerous changes can be made without defacing the original beneath. At the Patuxent Research

Refuge, the matte finish was found excellent for covering aerial photos, eliminating the glare from glossy prints without appreciably obscuring the details. When these photos were used for mapping vegetation, probable cover-types were speedily delineated in the laboratory, using an ordinary pencil. Then a relatively small amount of field work, using a red lead, sufficed to confirm or amend the boundaries and to delimit sub-types indistinguishable on the photographs.

When one area is under continuous scientific study, as at the Patuxent Research Refuge, wildlife census data or other observations can be recorded in the field on a map thus protected. After being copied and/or tabulated, the data can be erased leaving the map ready for use again.

Brief preliminary tests indicate many other uses for this material. For instance, the clear variety cut in strips can be used as a herbarium mounting tape in conjunction with the usual type of tape. Not requiring moistening and being transparent, it seems admirable for securing the more delicate plant parts. Its initial adhesion is not adequate to hold the more refractory parts, but where it does stick, it is claimed to form a permanent bond with the paper. Also, in the classroom, the sheeting can be used as a protective covering for illustrative material handed around for student use.

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## DISCUSSION

### THE BUSH REPORT AND SENATE BILLS

IN compliance with a request from President Roosevelt, dated November 17, 1944, Dr. Vannevar Bush undertook to advise the Government how science might be organized to serve our country in future days of peace as effectively as it was serving our armed forces during the war. In particular, the President inquired (a) what could be done, consistent with military security, to make known to the world the contributions to scientific knowledge during the war; (b) how a program might be organized to continue the scientific advances that had been made in the fields of medicine; (c) what the Government might at once and in the future do to aid research by public and private organizations; and (d) whether an effective program might be set up for discovering and developing scientific talent in American youth.

Dr. Bush appointed large committees of distinguished men to study and report on the four principal subjects enumerated above. After months of labor these committees presented comprehensive re-

ports, on the basis of which Dr. Bush made his report in compliance with the President's request. The report is developed under six chapter headings as follows: I, Introduction, a statement of the reasons that progress in science is essential and the relations of the Government to it; II, The War Against Disease; III, Science and the Public Welfare; IV, Renewal of Scientific Talent; V, A Problem of Scientific Reconversion; and VI, The Means to the End.

Under Chapter VI the report proposes a "National Research Foundation," including its purposes, its membership, its organization, its functions and duties, its patent policy, provisions for special authority, and its budget for five years. In short, it is a report that states a general problem of great importance, dissects it into its major components, sets up principles and outlines machinery for its administration, and proposes a financial budget, all fortified by comprehensive analyses and supporting data by very competent committees.

In spite of the excellence of the Bush Report, spe-