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CHARLES M. PROCTOR

Sanitary Engineering Center, Public Health Service, Cincinnati, Ohio

Meetings

Experimental "Allergic"

Encephalomyelitis

Since the development of Freund's adjuvants, much work has been done in experimental animals with nervous system antigens in attempts to elucidate the etiology and pathogenesis of various neurological disorders encountered in clinical and veterinary medicine. Many different disciplines have been involved—biochemistry, immunology, pathology, microbiology, and so on—so that the published reports are widely scattered in the world literature, in journals as well as in books. Much information pertaining to "allergic" encephalomyelitis has been published under titles which might not be recognized by either indexer or researcher as being related to this important experimental disease. In an attempt to assemble this large mass of pertinent data in a unified form, a symposium was held on "Experimental 'Allergic' Encephalomyelitis and Its Relation to Other Diseases of Man and Animals," 19 and 20 Oct. 1957, under the auspices of the National Advisory Council of the National Institute of Neurological Diseases and Blindness. Sixty scientists from many parts of the United States, Canada, England, Germany, France, Italy, and Japan met at the National Institutes of Health, Bethesda, Maryland, to discuss histologic, immunologic, and chemical aspects of these disorders. A brief summary of the data presented at this symposium may be of interest.

Experimental "allergic" encephalomyelitis can be produced in many species by the injection of vaccines containing brain and adjuvants, following which various clinical neurological signs develop, especially paralysis. A perivascular inflammation, often with demyelination, is seen histologically scattered through the central nervous system. Definition of the experimental disease requires consideration of (i) genetic and nutritional factors in the test animals; (ii) the use of "priming" injections of suspensions of *Hemophilus pertussis*; (iii) adjuvant factors and the route of inoculation; (iv) the chemical and immunologic nature of encephalitogenic materials isolated from neural or other tissues; (v) local and general reactions produced in the test animal; and (vi) the nature of the reaction within the nervous system.

H. A. Schneider (New York) summarized data obtained in mice which indicate that susceptibility to the experimental disease is inherited through a recessive gene. Susceptibility is influenced by other factors, however, and can be abolished by feeding genetically susceptible mice a synthetic diet which

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is adequate for maintenance of growth, especially if supplemented by Terramycin. Supplementation with vitamin B₁₂, folic acid, and biotin partially restores the susceptibility. Much further work must be done to define the crucial factors in the host and the environment, but it is possible that genetic and nutritional factors might account for the variable susceptibility of other species to different encephalitogenic materials.

Previous injection of suspensions of *H. pertussis* enhances the susceptibility of mice but has not been tried in other species. Its mechanism of action remains to be determined.

M. M. Lipton (Louisville, Ky.) reviewed the general mechanism of action of the components of Freund's adjuvants and concluded that the acid-fast bacilli and oil work together in the production of specific types of cells, directing the path of antibody formation toward cell-fixed as well as circulating types. The bacilli possibly also act as a *Schlepper*, making a hapten into a complete antigen. Of various routes, intradermal inoculation is generally considered most effective.

E. Lederer (Paris) correlated the immunologic and biologic effects of various lipids isolated from the tubercle bacillus: tubercle formation is produced by many branched-chain fatty acids (including mycolic acid); delayed sensitivity to another substance is induced by carbohydrate esters of mycolic acid; and adjuvant activity is related to wax-D Canetti (a tripeptide with mycolic acid and polysaccharide). He attributed J. Colover's (Taplow, England) production of encephalitis with brain and a protein residue of tubercle bacilli to an insoluble polymer of wax-D, possibly present in the bacterial wall. Further work is necessary to determine if large doses of tubercle bacilli might be inhibitory or if an optimal ratio exists between concentrations of the bacillary factors and the encephalitogenic "antigens."

Three types of chemical substances obtained from nervous tissue have been found to be encephalitogenic, but it is still not possible to account for the total activity of whole brain:

1) Proteolipids, in doses of about 35 mg, have been found by J. Folch, M. B. Lees, B. H. Waksman, and R. D. Adams (Boston) to be capable of producing a relatively mild form of the disease in rabbits; others have found proteolipids to possess only minimal activity in guinea pigs [G. Clark (Buffalo); Kies and Alvord] and none in rats [P. Y. Paterson (New York)]. Most of this activity resides in the "ether-soluble lower phase." The exact chemical composition of this fraction is at present not known, but it is known that it consists mainly of lipids with a small amount of protein (4 percent), presumably existing in proteolipid combination. J. M. Lee (New York) be-

lieves that proteolipid A, especially from homologous brain, is the only effective substance in mice.

2) Several proteins have been found by Kies, Roboz, and Alvord to be very effective in doses of 5 to 25 micrograms in guinea pigs, but others have found them not active in rats (Paterson), mice (Lee), and rabbits (Waksman). One of these proteins has been shown to be a single substance, by ultracentrifugation and electrophoresis, and to resemble a fragment of collagen in its high content of hydroxyproline and hydroxylysine (Roboz). Mild acid-extraction has yielded another protein-like encephalitogenic fraction from homologous guinea-pig brain (Kies).

3) A component has been isolated by Lipton from a petroleum ether extract

of brain. In amounts of only a few micrograms it is effective in guinea pigs. Its chemical nature remains to be identified. Lipton believes it to be a phospholipid, but there is enough nitrogen to account for half of it being protein.

Immunologic data concerning these substances are still fragmentary. It is not known how they relate to the species-specific protein-like, and species-nonspecific lipid-like, neural antigens which E. Witebsky (Buffalo) has shown to be immunologically related but different in brain, spinal cord, posterior pituitary, and adrenal medulla. The last two organs are not encephalitogenic. Preliminary experiments reported by Witebsky, in which Coons's fluorescent-labeled antibody technique was used, suggest their localization in myelin sheaths. Waksman

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reported delayed skin sensitization to homologous whole spinal cord or heterologous proteolipids paralleling the development of encephalomyelitis in rabbits, but M. W. Chase (New York) could not demonstrate such reactions to homologous brain in guinea pigs. M. Vulpé (Saskatoon, Canada) reported that a band appears between the alpha-1 and alpha-2 globulins in the electrophoretic pattern of serum of rabbits about 2 days before onset of paralysis, but whether this represents circulating antibody remains to be determined. Vulpé, A. Allegranza (Milan), and Murphy and Kies have failed to note any significant serum protein changes in guinea pigs during disease development.

B. Campbell and R. M. Condie (Minneapolis) emphasized the reaction of plasma cells not only locally and in the draining lymph nodes but also throughout the reticuloendothelial system and in the perivascular lesions in the central nervous system. In this last site, Adams and other neuropathologists insisted that adventitial histiocytes were also prominent. Correlation of the reaction of local epithelioid and distant cells with the various chemical fractions of the adjuvants and of brain remains to be made. Vogel's evidence on increased lipase activity in the regional lymph nodes has been extended by Vulpé and J. Olszewski, who found the degree of activity of this enzyme to be related to the lipid concentration of the material injected rather than to its encephalitogenic activity.

The site of the earliest lesion within the nervous system is still not determined: are the blood vessels (endothelium or adventitia) or the neural tissue (ground substance, glia, or myelin sheaths) primarily affected? Olszewski occasionally was able to demonstrate increased vascular permeability to radioactive iodinated serum albumin without leucocytic infiltration. L. Roizin (New York) has noted sudanophilic material in the endothelium, as well as marked accumulation of enzymes (oxidase, peroxidase, and phosphatase) with the perivascular cellular inflammation. He has also found periodic-acid-Schiff-positive, metachromatic material and sudanophilic, birefringent changes in myelin sheaths without inflammation.

The probable relation of the experimental disease to spontaneous neurologic diseases of man was discussed by H. Shiraki (Tokyo), who has studied the reaction occurring in human beings following injections of brain-containing vaccines used in the prevention of rabies. He described the differences between acute spinal and subacute or chronic cerebral forms of the reaction. Especially in the latter, large periventricular plaques strongly resembling multiple sclerosis plaques were noted.

The relation of experimental "aller-

gic" encephalomyelitis to multiple sclerosis and other demyelinating diseases in man was further considered by Adams, who felt that the basic reaction was a perivenous necrobiosis related to white matter and that it was more consistent with the hypothesis of an allergic reaction than with any other hypothesis. J. G. Greenfield (London), E. W. Hurst (Macclesfield, England), W. Haymaker (Washington, D.C.), A. Wolf (New York), A. Ferraro (New York), K. H. Finley (San Francisco), and J. R. M. Innes (Tuckahoe, N.Y.) generally agreed with Adams, but H. M. Zimmerman (New York) insisted that the nervous system can react only in a limited number of ways to any noxious agent. F. C. Robbins (Cleveland) mentioned that Russian investigators had isolated a virus from cases of acute multiple sclerosis and were experimenting with a vaccine.

Certain diseases of other organs (peripheral nerves, adrenal, testis, thyroid, lens, uveal tract, and various components of blood) have also been thought to be due to an allergic reaction, more particularly to an autoimmunization. Colover described his work on the adrenal, and Witebsky summarized data on the thyroid which strongly suggest that at least certain cases of chronic thyroiditis in man are due to the development of antibodies against the individual's own thyroglobulin. The only missing steps in the proof of this hypothesis are the production of thyroiditis by passive transfer of antibodies and the determination of the "trigger" mechanisms for release of thyroglobulin in man.

In discussing the pathogenesis of post-infectious encephalomyelitis, Robbins said he felt that the primary virus or another virus (latent or simultaneously infecting the patient) might directly invade the nervous system, break down the blood-brain barrier, and damage myelin or some other element of the nervous system. If the breakdown product then gets into the blood stream and becomes antigenic, the antibodies might in turn cause further damage in nervous tissue. Although W. S. Wood (Chicago) has found skin-sensitization to rabies vaccine in cases of rabies postvaccinal encephalomyelitis, he has not found it in cases of postinfectious encephalomyelitis.

Although all the available evidence concerning experimental "allergic" encephalomyelitis is consistent with the theory of allergy, it must be pointed out that at least two critical steps remain to be demonstrated: (i) the acceleration of the disease by previous specific sensitization, and (ii) the production of the disease by passive transfer of specific antibodies. Only negative or equivocal evidence for the latter has so far been obtained (Chase, Waksman, Vulpé, Wood, and Condie), and the available evidence concerning the former indicates that protection rather than acceleration

is afforded by prior injections of incomplete vaccines containing brain or adjuvants alone (Kies *et al.*, Paterson, Condie, Waksman, Zeman, and Ferraro). Although negative, these observations suggest that allergy may not play an important role in the development of the experimental disease and indicate that further work is necessary to settle an important question: is allergy a significant factor in diseases of the nervous system?

ELLSWORTH C. ALVORD, JR.
Baylor University College of
Medicine, Houston, Texas

MARIAN W. KIES
Laboratory of Clinical Science,
National Institute of Mental Health,
Bethesda, Maryland

Forthcoming Events

June

9-11. American Assoc. of Spectrographers, 9th annual symp., Chicago, Ill. (H. J. Hettel, Armour Research Foundation, 10 W. 35 St., Chicago 16.)

9-11. Canadian Federation of Biological Societies, 1st annual; with Canadian Assoc. of Anatomists, Canadian Biochemical Soc., Canadian Physiological Soc., and Pharmacological Soc. of Canada; Kingston, Ontario. (E. H. Bensley, Montreal General Hospital, 1650 Cedar Ave., Montreal 25, P.Q.)

9-11. Health Physics Soc., 3rd annual, Berkeley, Calif. (E. E. Anderson, Oak Ridge National Lab., Oak Ridge, Tenn.)

9-11. Soc. of General Physiologists, Woods Hole, Mass. (F. G. Sherman, Dept. of Biology, Brown Univ., Providence 12, R.I.)

9-11. Society for the Study of Development and Growth, 17th annual symp., South Hadley, Mass. (Miss K. Stein, Dept. of Zoology, Mount Holyoke College, South Hadley.)

9-12. Microscopy Symposium, 5th, Chicago, Ill. (W. C. McCrone, Jr., 500 E. 33 St., Chicago 16.)

9-13. Automation Exposition and Cong., 4th Internatl., New York. (International Automation Exposition, c/o Richard Rimbach Assoc., 845 Ridge Ave., Pittsburgh 12, Pa.)

10-12. Astronomical Soc. of the Pacific, annual, Los Angeles, Calif. (S. Einarsson, Leuschner Observatory, Univ. of California, Berkeley 4.)

10-13. Vacuum Techniques, 1st internatl. congress, Namur, Belgium. (E. Thomas, c/o CSN/ERM, 30, avenue de la Renaissance, Brussels 4, Belgium.)

11-14. Applied Mechanics, 3rd natl. Cong., Providence, R.I. (W. Prager, Brown Univ., Providence 12.)

11-14. National Soc. of Professional Engineers, St. Louis, Mo. (P. H. Robbins, NSPE, 2029 K St., NW, Washington, D.C.)

14-21. American Soc. of Medical Technologists, annual, Milwaukee, Wis. (Miss R. Matthaei, Suite 25, Hermann Professional Bldg., Houston 25, Tex.)

15-19. American Soc. of Mechanical



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Engineers, semiannual, Detroit, Mich. (O. B. Schier, II, ASME, 29 W. 39 St., New York 18.)

15-19. Cancer Research Conf., 3rd Canadian, Honey Harbour, Ontario. (R. L. Noble, Collip Medical Research Lab., Univ. of Western Ontario, London, Ont., Canada)

15-20. American Physical Therapy Assoc., annual, Seattle, Wash. (Miss M. E. Haskell, APTA, 1790 Broadway, New York 19.)

16-18. American Neurological Assoc., 83rd annual, Atlantic City, N.J. (C. Rupp, 133 S. 36 St., Philadelphia 4, Pa.)

16-18. Military Electronics Conv., 2nd,

Washington, D.C. (G. Rappaport, Emerson Radio & Phonograph Corp., 1140 East-West Highway, Silver Spring, Md.)

16-18. Photochemical Apparatus Symp., Upton, N.Y. (R. C. Fuller, Biology Dept., Brookhaven National Laboratory, Upton, L.I.)

16-20. American Soc. for Engineering Education, annual, Berkeley, Calif. (W. L. Collins, Univ. of Illinois, Urbana.)

16-20. Association of Official Seed Analysts, annual, Montreal, Quebec, Canada. (L. C. Shenberger, Seed Lab., Dept. of Agricultural Chemistry, Purdue Univ., Lafayette, Ind.)

16-20. Molecular Structure and Spec-

troscopy Symp., Columbus, Ohio. (R. A. Oetjen, Dept. of Physics and Astronomy, Ohio State Univ., Columbus 10.)

16-20. Pacific Div., AAAS, annual, Logan, Utah. (R. C. Miller, California Acad. of Sciences, Golden Gate Park, San Francisco 18.)

17-19. American Dairy Science Assoc., annual, Raleigh, N.C. (H. F. Judkins, 32 Ridgeway Circle, White Plains, N.Y.)

17-19. American Meteorological Soc., with Pacific Div., AAAS, Logan, Utah. (K. C. Spengler, AMS, 3 Joy St., Boston 8, Mass.)

18-20. Statistical Methods in Radio Wave Propagation, intern. symp., Los Angeles, Calif. (W. C. Hoffman, 3116 Engineering Bldg., Univ. of California, Los Angeles 24.)

18-21. College Physicists, 20th annual colloquium, Iowa City, Iowa. (J. A. Van Allen, Dept. of Physics, State Univ. of Iowa, Iowa City.)

18-22. American College of Chest Physicians, annual, San Francisco, Calif. (M. Kornfeld, ACCP, 112 E. Chestnut St., Chicago 11, Ill.)

19-21. Endocrine Soc., 40th annual, San Francisco, Calif. (H. H. Turner, 1200 N. Walker St., Oklahoma City 3, Okla.)

19-21. Society of Nuclear Medicine, 5th annual, Los Angeles, Calif. (R. W. Lackey, 452 Metropolitan Bldg., Denver, Colo.)

19-25. Scandinavian-American Meteorological Meeting, Bergen, Norway. (K. C. Spengler, 3 Joy St., Boston, Mass.)

21-22. Society for Investigative Dermatology, annual, San Francisco, Calif. (H. Beerman, 255 S. 17 St., Philadelphia 3, Pa.)

22-25. American Soc. of Agricultural Engineers, 51st annual, Santa Barbara, Calif. (J. L. Butt, ASAE, St. Joseph, Mich.)

22-25. Medicinal Chemistry, 6th natl. symp., Madison, Wis. (E. Smismann, College of Pharmacy, Univ. of Wisconsin, Madison.)

22-27. American Inst. of Chemical Engineers, 50th anniversary, Philadelphia, Pa. (F. J. Van Antwerpen, AIChE, 25 W. 45 St., New York 36.)

22-27. American Soc. for Testing Materials, 61st annual, Boston, Mass. (F. F. Van Atta, ASTM, 1916 Race St., Philadelphia 3, Pa.)

23-24. Unstable Chemical Species Symp., Los Angeles, Calif. (Directorate of Advanced Studies, Air Force Office of Scientific Research, P. O. Box 2035-D, Pasadena, Calif.)

23-25. American Soc. of Heating and Air-Conditioning Engineers, semiannual, Minneapolis, Minn. (A. V. Hutchinson, ASHAE, 62 Worth St., New York 13.)

23-25. American Soc. of Refrigerating Engineers, annual, Minneapolis, Minn. (R. C. Cross, ASRE, 234 Fifth Ave., New York 1.)

23-27. American Soc. of Civil Engineers, Portland, Ore. (W. H. Wisely, ASCE, 33 W. 39 St., New York 18.)

23-28. Low Temperature Physics, 6th internatl. conf., Leiden, Netherlands. (J. van den Handel, Kamerlingh Onnes Laboratory, Leiden.)

(See issue of 18 April for comprehensive list)

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