

does parallel, however, within limits, the increased weight of the lung which takes place during infection with influenza virus. Third, in certain fractionation-experiments reactions of a somewhat different character have been observed in the absence of virus.

It may be that the antigen is a combination of active and altered virus, and that infectivity for mice is not an accurate measurement of the amount of virus that can react with the serum. On the other hand, it may be that the flocculating antigen is some substance other than the virus which appears in the mouse-lung during infection, similar perhaps to the nonspecific precipitinogen in yellow fever described by Hughes.³ The problem is being investigated further.

10099

Effect of Sulfanilamide upon the Viability of Meningococci in Spinal Fluid.

ERWIN NETER.

From the Laboratories of the Children's Hospital and the Department of Pathology and Bacteriology, University of Buffalo, School of Medicine.

The investigations of Branham and Rosenthal,¹ Buttle, Gray, and Stephenson,² McPherson Brown,³ Proom,⁴ Rosenthal, Bauer, and Branham,⁵ have established the curative value of sulfanilamide and related compounds in experimental meningococcal infections of animals. In man, favorable results in the treatment of meningococcal meningitis by means of sulfanilamide (either alone or in conjunction with antimeningococcal serum) were reported by numerous observers.⁶⁻¹⁷ In a recent communication,¹⁸ it was reported that sul-

³ Hughes, T. P., *J. Immunol.*, 1933, **25**, 275.

¹ Branham, S. E., and Rosenthal, S. M., *Publ. Health Rep.*, 1937, **52**, 685.

² Buttle, G. A., Gray, W. R., and Stephenson, D., *Lancet*, 1936, 1286.

³ McPherson Brown, Th., *Bull. Johns Hopkins Hosp.*, 1937, **61**, 272.

⁴ Proom, H., *Lancet*, 1937, 16.

⁵ Rosenthal, S. M., Bauer, H., and Branham, S. E., *Publ. Health Rep.*, 1937, **52**, 662.

⁶ Schmidt, W., *Deutsche med. Wchnschr.*, 1936, **62**, 881.

⁷ Schwentker, F. F., Gelman, S., and Long, P. H., *J. Am. Med. Assn.*, 1937, **108**, 1407.

⁸ McIntosh, R., Wilcox, D. A., and Wright, F. H., *J. Pediat.*, 1937, **11**, 167.

⁹ Mitchell, A. G., and Trachsler, W. H., *J. Pediat.*, 1937, **11**, 183.

¹⁰ Brennemann, J., *J. Pediat.*, 1937, **11**, 238.

¹¹ Bernstein, S. S., *J. Pediat.*, 1937, **11**, 198.

fanilamide may exert a bacteriostatic action upon meningococci present in spinal fluid of patients with meningococcal meningitis. When such spinal fluids were incubated with sulfanilamide, growth of meningococci failed to take place, while the number of microorganisms in a specimen without sulfanilamide showed a marked increase. In the following, experiments are being reported dealing with the action of sulfanilamide upon the viability of meningococci.

The experiments were carried out in the following manner: Spinal fluid was obtained from patients with meningococcal meningitis prior to treatment. It was taken and kept under sterile precautions. Ten drops of spinal fluid were mixed with equal amounts of sulfanilamide in various concentrations, ranging from 0.8% to 0.0064%. The preparation used was Prontylin, Winthrop, repurified for injection, and was dissolved in physiological saline solution. The spinal fluid-sulfanilamide mixtures were then incubated at 37°C. After various periods of incubation, subcultures were made on 30% ascitic-chocolate-agar plates. The plates were incubated at 37°C in a jar containing about 10% CO₂. The resulting growth of meningococci was noted. Table I gives the results of such an experiment.

TABLE I.
Action of Sulfanilamide upon Viability of Meningococci in Spinal Fluid.

Concentration of sulfanilamide %		Growth of meningococci on ascitic-chocolate-agar at 37°C after treatment with sulfanilamide for—							
		A 5 min		B 45 min		C 3 hr		D 24 hr	
		a	b	a	b	a	b	a	b
1.	0.4	++	++	+	++	0	+	0	0
2.	0.08	++	++	++	++	0	++	0	0
3.	0.016	++	++	++	++	++	++	0	0
4.	0.0032	++	++	++	++	++	++	+	++
5.	0	++	++	++	++	++	++	++	++

a After 18 hr at 37°C.

b After 48 hr at 37°C.

++ Marked growth

+ Slight growth.

0 No growth.

It may be seen from this table, that treatment for 5 minutes of meningococci in spinal fluid, with various concentrations of sulfanila-

¹² Basman, J., and Perley, A. M., *J. Pediat.*, 1937, **11**, 212.

¹³ Carey, B. W., *J. Pediat.*, 1937, **11**, 202.

¹⁴ Zedel, J. F., and Greenberg, D., *New York State J. Med.*, 1937, **37**, 1744.

¹⁵ Willien, L. J., *J. Am. Med. Assn.*, 1938, **110**, 630.

¹⁶ Eldahl, A., *Lancet*, 1938, 712.

¹⁷ Crawford, T., and Fleming, G. B., *Lancet*, 1938, 987.

¹⁸ Neter, E., *PROC. SOC. EXP. BIOL. AND MED.*, 1938, **38**, 37.

mide did not inhibit the growth of this microorganism. When the treatment with sulfanilamide was carried out for 3 hours, the growth of meningococci on subculture was retarded. While growth of meningococci from spinal fluid containing 0.016% sulfanilamide, or less, occurred after 18 hours' incubation at 37°C, growth from spinal fluid containing 0.08% sulfanilamide, or more, appeared only after 48 hours' incubation. On the other hand, when spinal fluid was treated with sulfanilamide in concentrations of 0.016% or more for 24 hours, no growth was obtained on subculture. Substantially the same results were obtained with spinal fluids of 3 other patients with meningococcal meningitis.

It follows from these experiments that the effect of sulfanilamide upon the viability of meningococci in spinal fluid depends upon the concentration of the drug as well as upon the period allowed for its action. The experiments furthermore show that the small quantities of sulfanilamide, present in the spinal fluid-sulfanilamide mixtures used for the inoculation of the medium cannot be responsible for the delay and inhibition of growth, since meningococci grew profusely within 18 hours from spinal fluid-sulfanilamide mixtures being kept for short periods only.

In order to elucidate whether the above described effects of sulfanilamide upon meningococci present in spinal fluids of patients with meningococcal meningitis depends upon the presence of leukocytes, parallel experiments were carried out with meningococci suspended in infusion-broth. It was found that treatment with sulfanilamide may result in loss of viability of meningococci in the absence of leukocytes.

In summary, sulfanilamide may have the following action upon meningococci: Firstly, in the presence of sulfanilamide, the growth of meningococci may be inhibited. Secondly, previous treatment of meningococci with sulfanilamide may result in the retardation of their growth on subculture or in the loss of their viability, depending upon concentration of the drug as well as upon the period allowed for its action.