10824

Penetration of Antiserum into the Central Nervous System of Monkeys Infected with Poliomyelitis.* II.

J. Emerson Kempf and Malcolm H. Soule.

From the Hygienic Laboratory, University of Michigan, Ann Arbor.

It has been shown in the monkey infected with poliomyelitis that foreign serum injected intravenously cannot be detected in significant amounts in the cerebrospinal fluid nor in the CNS tissue.[1, 2, 3] Other investigators,[4-7] using a variety of antibodies in normal and diseased conditions have noted similar impermeability of the blood-brain barrier.

Netter and Debré[8] in 10 cases of meningitis noted the presence of horse serum in the blood of 8 patients less than 20 minutes after intrathecal injection. Stern and Gautier[5] introduced dyes and antiserum into the spinal fluid and readily detected them in the blood, while the same substances when given intravenously did not appear in significant quantities in the cerebrospinal fluid. McKinley and Holden[9] found *B. coli* "D" phage filterable from the cerebrospinal fluid into the blood, but not in the reverse direction. Burtenshaw[4] following intraspinal injection of horse antimeningococcus serum observed that it rapidly decreased in quantity in the spinal fluid and quickly appeared in the blood. Within one or two days the cerebrospinal fluid content fell to the low levels attained by parenteral administration alone.

Friedemann, Zuger and Hollander[10] believed that the cerebral capillaries of the guinea pig and rabbit are permeable to antibodies, because in their experiments tetanus antitoxin given intravenously

* This work was aided by a grant from the Clara Ward Seabury Clinic for Infantile Paralysis.

1 Shaughnessy, H. S., Grubb, T. C., and Harmon, P. H., *J. Bact.*, 1937, **33**, 59.

2 Lennette, E. H., and Campbell, D. H., Proc. Soc. Exp. Biol. and Med., 1939, **41**, 320.

3 Kempf, J. E., Nungester, W. J., and Soule, M. H., Proc. Soc. Exp. Biol. and Med., 1939, **40**, 395.

4 Burtenshaw, J. M. L., *Lancet*, 1938, **2**, 1513.

5 Stern, L., and Gautier, R., *Arch. Intern. Physiol.*, 1921, **17**, 138.

6 Katzenelbogen, S., and Goldsmith, H., *Am. J. Psychol.*, 1931, **10**, 1045.

7 Hektoen, L., and Carlson, J., *J. Infect. Dis.*, 1910, **7**, 319.

8 Netter, A., and Debré, R., *Comp. Rend. Soc. de Biol.*, 1909, **67**, 100.

9 McKinley, E. B., and Holden, W., Proc. Soc. Exp. Biol. and Med., 1927, **24**, 595.

10 Friedemann, U., Zuger, B., and Hollander, A., *J. Immunol.*, 1939, **36**, 219.

neutralized tetanus toxin injected directly into the ventricles. On the other hand, Ramon and Descombey[11] produced a fatal tetanus in guinea pigs with blood from rabbits which had a few hours previously received intraspinal tetanus toxin. It would seem that Friedemann's, Zuger's and Hollander's results may be explained by assuming that the toxin injected into the cerebrospinal fluid passed into the blood stream and was there neutralized by the circulating antibodies. Since tetanus symptoms did not develop in their untreated animals until approximately 7 days after inoculation, toxin was not in the nerve tissue in significant quantities but may have been circulating in the blood in accordance with the work of Abel.[12] Moreover, mechanical rupture of blood vessels and resultant inflammatory reactions following intracranial inoculation might afford another direct route for the passage of toxin into the blood.

It is widely believed that antiserum injected intrathecally penetrates the CNS tissue. However, Flexner and Lewis[13] found it difficult to neutralize poliomyelitis virus suspensions in amounts as small as 1/50 cc when they were administered intracranially and the antiserums were given intrathecally at the same time. Cestan, Riser, and Labordee,[14] on injecting ferrocyanide into the ventricles during life and examining for Prussian blue granules post mortem, detected none deep in the nerve tissue. On such a basis they concluded that the cerebrospinal fluid does not penetrate the nerve cell, and that the intrathecal method of therapy is of no avail.

In the present work an attempt was made to demonstrate the penetration into the CNS tissue of rabbit hemolysin serum against sheep red blood cells following cisternal injection in animals during the active stage of poliomyelitis. Monkeys previously inoculated intracranially with Aycock's strain of poliomyelitis virus were given cisternally one to 2 cc of rabbit hemolytic serum with a titer varying from 1:30,000 to 1:40,000. After a definite time interval had elapsed (Table I), the animals were sacrificed. Spinal puncture was then performed, and specimens of spinal fluid were withdrawn. Subsequently, various portions of the brain and spinal cord were removed, and approximately 0.5 g samples mixed with physiological saline solution and ground with sand. The resulting suspensions were centrifuged and the supernatant fluid was tested for hemolysins according to the technic described in a previous paper.[8]

11 Ramon, G., and Descombey, P., *Comp. Rend. Soc. de Biol.*, 1931, **108**, 358.

12 Abel, J. J., *Science*, 1934, **79**, 63.

13 Flexner, S., and Lewis, P. M., *J. A. M. A.*, 1910, **54**, 178.

14 Cestan, Riser, Labordee, *Medicine*, Paris, 1926, **7**, 1931.

TABLE I.
Penetration of Hemolysin in Nerve Tissue of Monkeys with Poliomyelitis.

Monkey	Stage of disease when hemolysin was injected	Hr between injection and sacrifice	Titer of hemolysin in blood	Titer of hemolysin shown as % of corresponding blood sample*					
				Spinal fluid	Lumbar cord	Cervical cord	Medulla	Site of inoculation of lesion	Frontal lobe
6	Quadriplegia	12	1:320	100	0	0	0	0	0
7	"	6	1:640	6.2	0	0	0	12	0
8	"	7	1:640	0	0	0	0	0	0
9	"	10	1:1280	50	0	0	0	0	0
10	"	12	1:2560	3	1.5	1.5	3	1.5	1.5

*The lowest dilution of tissue not anti-complementary was 1:40.

In a period of 12 hours or less (Table I) hemolysin could be detected in the blood in a minimum titer of 1:640 in 4 out of 5 cases. A titer as low as 1:320 was noted in only one instance. The hemolytic titers in the cerebrospinal fluid were definitely lower than the titers of the blood. The CNS tissues in most instances showed no hemolysin, the lowest dilution examined being 1:40. In one sample of brain taken from the site of inoculation of the virus, the hemolysin titer was 12% of the blood value.

These findings demonstrate that practically none of the hemolysin administered into the cerebrospinal fluid in monkeys infected with poliomyelitis actually reaches the CNS. The antibodies introduced left the cerebrospinal fluid within the experimental time of 6 to 12 hours and were demonstrable in high titer in the circulating blood.

Shaughnessy[15] has made similar observations.

10825

Effects of Activity upon Tissues of the Rat.

J. T. McCLINTOCK, H. M. HINES AND DOROTHY P. JORDAN.

From the Department of Physiology, College of Medicine, State University of Iowa, Iowa City.

This investigation was undertaken in order to determine the effect of different degrees of activity upon the weight and composition of tissues of growing animals. It was thought that this information might throw some light upon the mode of adjustment of the organism to the effects of chronic exercise and aid in the interpretation of observations made upon animals in which muscular activity was a variable factor. In this report are included observations of body and heart weights and the concentration of creatine, hemoglobin, total nitrogen and water in the skeletal muscle of 2 groups of albino rats. Litter mates after being weaned at the age of 5 weeks were equally distributed between 2 groups. The activity of one group, designated as inactive, was restricted by confining the animals to small individual cages for a period of 6½ months. The cages measured 21 cm in diameter by 22.5 cm in height and were almost completely filled with cut paper to further restrict movement. The other group, referred to as active, were housed in revolving activity cages.[1] Food and water were allowed *ad libitum*. The animals were

[15] Shaughnessy, H. J., personal communication.

[1] Durrant, E. P., *Am. J. Physiol.*, 1924, **70**, 344.