TABLE I.
Wheat Germ Oil Experiments. Autopsy Findings Normal.

No. of rats	Sex	Wheat germ oil: Type	Quantity fed 6 times weekly	As % of diet ad lib.	Days fed
20	F	petroleum ether extracted	2 cc		45
13	F	" " "		22 (Diet 789)	45
14	F	" " "		10 (" 791)	365
14	F	" " "	4 drops		365
5	M	" " "	4 "		277
12	F	pressed		50 (" 798)	180
6	M	"		50 (" 798)	180
6	F	vitamin E concentrate	100 mg		180
3	M	" " "	100 mg		180
6	F	ether extracted		30 (" 809)	370
2	M	ether extracted		30 (" 809)	370

The animals in all groups were autopsied at the end of the periods indicated (45-370 days) and a careful examination of all organs was made. In no case was a neoplasm found (Table I) even in the case of rats maintained on the 30% ether extracted wheat germ oil for a period of 370 days. It must be pointed out that our rats are not of the albino strain and may be sturdier than those employed by Rowntree.

Conclusion. Rats of the Long-Evans strain did not develop abdominal neoplasms when maintained for periods from 45-370 days on high wheat-germ-oil-containing rations. One hundred and one animals were employed in these experiments.

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Blood-CNS Barrier Permeability to Horse Serum In Experimental Poliomyelitis.*

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It is generally accepted that the passage of foreign substances between the blood and spinal fluid is controlled by a physiologic barrier interposed between the central nervous system and other,

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non-nervous tissues. This barrier, usually referred to as the blood-brain (or blood-CNS[1]) barrier, is regarded as a functional entity[2] composed of various structures of the cerebrospinal nervous system. Stern and her collaborators[3] have adduced evidence that certain portions of the barrier are concerned mainly with the passage of crystalloids while other structures are concerned with colloidal substances.

We have previously reported that the pathologic changes occurring in the CNS of *Rhesus* monkeys infected with poliomyelitis virus increase the permeability of the barrier to crystalloids such as sodium nitrate[4] and sodium bromide.[5] Since the penetration of crystalloids and colloids from the blood into the cerebrospinal fluid appears to be controlled by 2 different mechanisms, we believed the bearing of this subject on chemotherapy and serum therapy warranted further examination. While this work was in progress, Kempf, Nungester and Soule[6] reported that rabbit antisheep hemolysin passes the barrier in insignificant amounts; this experience agrees well with that of Shaughnessy, Grubb and Harmon,[7] who used several antibody-containing sera.

In these experiments we have used *Rhesus* monkeys in various stages of the disease following cerebral or nasal inoculation of the potent MV strain of virus. In order to maintain during the experiment approximately constant concentration of foreign protein in the bloodstream, normal horse serum† was administered subcutaneously in 5.0 cc amounts twice daily for 2 consecutive days. On the 3rd day, specimens of blood and spinal fluid for analysis were obtained by cardiac and cisternal puncture under deep ether anesthesia. Detection and rough estimation of the concentration of horse serum in the serum and spinal fluid of monkeys was accomplished by using standardized precipitating serum from rabbits which had been immunized with horse serum. The concentration of horse serum present was calculated from the "end-point" or limiting dilution

1 Lennette, E. H., and Hudson, N. P., PROC. SOC. EXP. BIOL. AND MED., 1936, **34**, 470.

2 Katzenelbogen, S., *The cerebrospinal fluid and its relation to the blood,* Baltimore, Johns Hopkins Press, 1935.

3 Stern, L., quoted from Katzenelbogen, *loc. cit.*, p. 83.

4 Lennette, E. H., and Reames, H. R., *J. Immunol.*, 1938, **34**, 215.

5 Lennette, E. H., and Campbell, D. H., *Am. J. Dis. Child.*, 1938, **56**, 756.

6 Kempf, J. E., Nungester, W. J., and Soule, M. H., PROC. SOC. EXP. BIOL. AND MED., 1939, **40**, 395.

7 Shaughnessy, H. J., Grubb, T. C., and Harmon, P. H., *J. Bact.*, 1937, **32**, 47.

† We are indebted to Parke, Davis and Co. for a generous supply of normal horse serum.

required to form a precipitate with the previously standardized antiserum. For example, if the limiting dilution of a sample of monkey serum containing horse serum was 1:600 when tested against antiserum having a titer of 1:100,000, the concentration of horse serum was expressed as $600/100{,}000 = 0.600\%$. The antiserum was standardized by titration against a known amount of horse serum added to monkey serum or spinal fluid. When testing an unknown specimen, a preliminary titration was first done to ascertain the approximate range and then a second titration was performed to secure a more accurate value. The final dilutions of spinal fluid tested were 1:2, 1:4, 1:6, 1:8, etc., and of serum 1:50 with each succeeding dilution increased by 20%. Although a few of the tests were carried out by the use of the "ring" method, the majority were done by mixing 0.3 cc amounts of the antiserum which had been diluted with one volume of 0.5% saline, with 0.3 cc of the test solution in small thin-walled tubes. The mixtures were incubated for 1 hour at 45°C, then placed in a refrigerator at 4°C, and read after 2 hours.

Since we[8] among others have noted that determination of the concentration of the test substance in the spinal fluid alone gives only a very rough idea of barrier permeability, the results are expressed here as a ratio,

$$\frac{\text{horse serum in blood}}{\text{horse serum in spinal fluid}},$$

which is referred to as the P.Q. (Permeability Quotient). The P.Q. values given in Table I represent the following ratio:

$$\frac{X_s/S}{X_{csf}/S}$$

where X_s represents the limiting dilution of the unknown sample of monkey serum, X_{csf} the limiting dilution of spinal fluid and S the titration value of the standard antiserum. This procedure gives a more precise idea of barrier permeability, and it should be noted that a high P.Q. value corresponds to a low degree of permeability, and that as permeability increases, the P.Q. value decreases.

From Table I it will be seen that we were unable to detect any horse serum in the spinal fluid of normal control monkeys and hence the P.Q. could not be calculated. On the other hand, there was some increased permeability to horse serum in the infected animals. The quotients obtained were much higher than those obtained with sodium bromide[5] and tend to approximate those obtained by Bur-

8 Lennette, E. H., Campbell, D. H., and Reames, H. R., PROC. SOC. EXP. BIOL. AND MED., 1939, **40**, 287.

TABLE I.
Permeability of Blood-CNS Barrier to Horse Serum in Poliomyelitis.

History of monkeys	No. of monkeys	Permeability quotients
Normals	5	0*, 0, 0, 0, 0
Preparalytic	3†	100, 120, 200
	4‡	20, 40, 160, 200
Paralytic	4†	160, 240, 340, 760
	4‡	20, 60, 120, 600

* No horse serum detected in spinal fluid.
† Poliomyelitis following nasal instillation of virus.
‡ " " intracerebral inoculation of virus.

tenshaw[9] in human meningococcus meningitis. The high P.Q. values indicate that horse serum penetrated the barrier in small and, from a therapeutic standpoint, probably insignificant amounts. This may be due in part to the fact that horse serum disappears from the bloodstream relatively rapidly and hence fails to maintain the concentration necessary for penetration of the material into the spinal fluid. Whether the same situation obtains with homologous protein is now under study.

Summary. Normal horse serum administered in the quantity and manner described was found to penetrate the blood-CNS barrier of poliomyelitic animals regularly although in small amounts. From a therapeutic standpoint, it would seem that under the stated conditions such concentrations of serum would probably be inadequate.

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Comparison of Activity of Viruses of St. Louis and Japanese Encephalitis in the Chick Embryo.

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The close similarity in the clinical and pathologic pictures of the St. Louis and the Japanese Type B encephalitis in both man and the mouse suggested a comparative study of the activity of these viruses in the chick embryo.

[9] Burtenshaw, J. M. L., *Lancet*, 1938, **2**, 1513.