with the stock Swiss mice showed incubation periods with approximately the same range of variation. (Table I). The one exception, the Rfi strain, also showed variable incubation periods but in a greater number of this group the period was exceedingly long. Eight of 12 mice of this strain died between 30 and 54 days after inoculation. Although some of the Swiss control mice used in the experiment with mice of the Rfi strain showed more prolonged incubation periods than the control mice in the other experiments, the average incubation period of the mice of the Rfi strain in comparison with that of the control mice appeared to be significantly longer (Table I).

It is also noteworthy that a greater proportion of mice of the C57 and dba strains became paralyzed and died during the first few days after inoculation. With the relatively small number of animals used there were no significant differences in the percentage of deaths among the several strains of mice.

Conclusions. It is concluded from these observations that, although strain differences are found, other factors than constitution are important in determining the variable incubation periods observed with the Lansing strain of poliomyelitis virus within strains of white mice.

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Demonstration of Murine Poliomyelitis Virus in Blood and Spleens of Mice Shortly Following Inoculation.*

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A few positive results have been obtained in attempts to demonstrate the virus of poliomyelitis (M V strain) in the blood of Rhesus monkeys. Clark, Fraser and Amoss[1] demonstrated the virus in the defibrinated blood of monkeys for 72 hr following the intracerebral and intraspinal inoculation of overwhelming doses of virus, and for 24 hr after the intravenous injection of the virus. Flexner and Lewis[2] produced the disease when large quantities of blood (25 cc), withdrawn from infected monkeys at the height of the disease, were injected intravenously. However by intravenous inoculation Gordon and Lennette[3] were unable to demonstrate the virus in large amounts of the blood of Rhesus monkeys obtained at various stages of the experimental disease. Others[1,4] have reported the occasional demonstration of the virus by the intracerebral injection of smaller amounts of defibrinated blood withdrawn from monkeys after the onset of paralysis. The virus of poliomyelitis was demonstrated inconstantly by Lennette[5] in the emulsions of perfused spleens removed from paralyzed monkeys which had been infected by the intracerebral route.

The demonstration of the Lansing strain of poliomyelitis virus in the blood or spleen of mice following the intraperitoneal or intracerebral inoculation of the virus has not been reported. The present studies were carried out to gain further knowledge of the behavior of this virus in mice.

Experimental. In these experiments the Lansing strain of poliomyelitis virus (Armstrong virus) was used. One group of 50 Swiss mice, 6 to 8 weeks of age, was inocu-

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1 Clark, P. F., Fraser, F. R., and Amoss, H. L., *J. Exp. Med.*, 1914, **19**, 223.

2 Flexner, S., and Lewis, P. A., *J. Exp. Med.*, 1910, **12**, 227.

3 Gordon, F. B., and Lennette, E. H., *J. Inf. Dis.*, 1939, **64**, 97.

4 Leiner, K., and Von Wiesner, R., *Wien. Klin. Wchnschr.*, 1909, **22**, 1698.

5 Lennette, E. H., *J. Exp. Med.*, 1937, **66**, 549.

TABLE I.
Demonstration of Virus in Blood and Spleens of Swiss Mice Following Intraperitoneal Inoculation of Lansing Strain of Poliomyelitis Virus.

Hours after intraper. inocul. of virus	Material tested: Whole blood 1	Whole blood 2	Pooled sera of 3 mice	Pooled emulsion of 3 spleens
½-¾	3/3*	—	3/3	3/6
1½	3/3	3/3	—	6/6
2½	2/3	0/3	0/3	6/6
5	3/3	2/3	1/3	4/5
7	0/3	0/3	2/3	3/3
12	0/3	0/3	0/2	2/4
24			No virus demonstrated	
72			,, ,, ,,	
96			,, ,, ,,	
120			,, ,, ,,	

*Denominator = No. of mice inoculated.
Numerator = No. of deaths attributable to the virus.

lated intraperitoneally, each receiving .3 cc of a 1:10 dilution in broth of infected mouse-brain and spinal cord. Another group of 45 mice was inoculated intracerebrally with .03 cc of a 1:5 dilution in broth of infected mouse-brain and spinal cord. The blood and spleens of the injected mice were tested for the presence of virus by the intracerebral inoculation of Swiss mice, 6 to 8 weeks of age. These materials were tested at 10 different periods from ½ hr to 120 hr following the intraperitoneal injection of the virus and at 9 periods from ½ hr to 72 hr following the intracerebral injection. At each time the whole blood of each of 2 mice and also the pooled sera of 3 mice were tested. Spleens of the 3 mice, from which the pooled sera were obtained, were emulsified together in broth, using ½ to ¾ cc of broth per spleen depending upon the size of the organ. The emulsions were allowed to settle without centrifugation and the supernatant fluid used for the intracerebral inoculation of 3 to 6 mice.

The whole blood, the pooled sera and the splenic emulsions contained virus at ½ hr and for as long as 5 hr following the intraperitoneal inoculation (Table I). At 7 hr the virus was demonstrated in the pooled sera and the splenic emulsion, and at 12 hr only in the splenic emulsion. Later than 12 hr following the intraperitoneal inoculation virus was not found in either the blood or spleen.

After the intracerebral injection (Table II) virus was demonstrated in the whole blood for ½ to 2½ hr and in the spleens only at 1½ hr following inoculation.

TABLE II.
Demonstration of Virus in Blood and Spleens of Swiss Mice Following Intracerebral Inoculation of Lansing Strain of Poliomyelitis Virus.

Hours after intracer. inocul. of virus	Material tested: Whole blood 1	Whole blood 2	Pooled serum	Pooled emulsion of 3 spleens
½-¾	1/3*	1/3	2/3	0/6
1½	2/3	2/3	1/2	1/6
2½	1/3	0/2	1/4	0/6
5			No virus demonstrated	
7			,, ,, ,,	
12			,, ,, ,,	
24			,, ,, ,,	
48			,, ,, ,,	
72			,, ,, ,,	

*Denominator = No. of mice inoculated.
Numerator = No. of deaths attributed to the virus.

The blood and spleens of 12 mice infected by the intracerebral route with the Lansing strain of poliomyelitis virus were tested for the presence of the virus after the mice became paralyzed, but no virus was demonstrated in either of these materials at that time.

Although the Lansing strain of poliomyelitis virus reaches the blood shortly after intraperitoneal and intracerebral inoculation of large doses of virus, it persists in the blood for only a short time. The time for which the virus persists in the spleen and in the blood is approximately the same. Therefore it is probable that the presence of the virus in the spleen depended upon the blood contained in that organ.

Summary. The Lansing strain of poliomyelitis virus has been demonstrated in the blood and in the spleen of mice ½ hr following the intraperitoneal inoculation of the virus. It has been demonstrated in the blood for 7 hr and in the spleen for 12 hr following intraperitoneal inoculation but not later. After intracerebral inoculation it has been demonstrated in the blood at intervals as long as 2½ hr but in the spleen only at 1½ hr following inoculation. The virus was not demonstrated in the spleens or blood of 12 mice when paralysis had developed following the intracerebral inoculation of the virus.

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In vitro Effects of Quinine, Atabrine and Substituted Acridine Compounds upon Gram Negative Bacteria.

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Since atabrine has been found to be a suitable chemotherapeutic agent for the treatment and prophylaxis of malaria[1,2] and is furthermore indicated in the treatment of giardiasis,[3,4] it appeared worthwhile to compare the *in vitro* effects of these compounds and additional substituted acridines upon other groups of microörganisms. This communication presents the effects of several such preparations upon organisms of the colon-typhoid-dysentery and related groups of gram-negative bacteria in broth.

The products tested included 3-chloro-7-methoxy-9-(1-methyl-4-diethylamino) butylamino-acridine-dihydrochloride (Atabrine); 3-chloro- 7-methoxy- 9-(2-hydroxy-3- diethylamino) propylamino-acridine-dihydrochloride (P-1); 3-chloro-7-methoxy-9-(2-hydroxy-ethylamino) ethylamino-acridine-dihydrochloride (P-2); 3-chloro-7-methoxy-9-(4-sulfamidobenzyl) acridine (P-3); 2-ethoxy-6,9-diamino-acridine (Rivanol); 2-ethoxy-6, 9-diamino-acridine lactate (Rivanol lactate); and 2-ethoxy-6, 9-diamino-acridine-azo-sulfanilamide (Rivanol-azo-sulfanilamide).* Quinine hydrochloride was included in these tests for comparative purposes.

Method. Dilute suspensions of 22-24 hr test cultures grown in beef extract broth were inoculated in 0.2 cc amounts in 5 cc of the broth medium containing 50 mg % concentration of one of the compounds. The inoculated drug-broth mixtures, together with control tubes lacking a drug, were incubated at 37° C. Presence or absence of visible growth was recorded at intervals of 24, 48 and 72 hr. Lack of visible growth at the end of 24 hr was taken as an indication of inhibition. In order to differentiate between bacteriostatic and possible bactericidal actions of the agents tested 1 cc of the test

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* The Rivanol-azo-sulfanilamide compound was kindly supplied by Dr. Frederick Proescher, San Jose, California.