Jersey" strain, and in one the tissue culture virus of the latter strain. Three typical experiments are shown in Tables I, II, and III. It will be seen from these tables that the virus passed through all membranes which had an average pore diameter of 150 mμ or greater. The passage through the 140 mμ membranes was irregular, as in some of the experiments the filtrates proved infective, while in others they failed to produce infection. The filtrates of membranes which had an average pore diameter of 130 or less gave uniformly negative results. The filtration end-point, therefore, is considered to be approximately 140 mμ. These results are in close agreement with those obtained by Galloway and Elford.

Summary. The filtration end-point of the virus of vesicular stomatitis, or the average pore diameter of the finest membrane passing the virus, was found to be approximately 140 mμ. Two immunologically distinct strains of the virus, the "Indiana" and the "New Jersey" maintained either in tissue culture or in mouse brain, were studied, and the filtration end-point was found to be the same irrespective of the source or serological type of the virus.

Our results confirm those of Galloway and Elford, who found that the virus passes through collodion membranes which have an average pore diameter of 160 mμ but is held back by those of 130 mμ. From their results they estimated the particle size of the virus to be between 70 and 100 mμ.

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Early Diagnosis of Rabies by Mouse Inoculation. Measurement of Humoral Immunity to Rabies by Mouse Protection Test.

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A method for early and reliable diagnosis of rabies by animal inoculation and a protection test for measuring humoral immunity to the virus may become feasible by the use of highly susceptible strains of mice. Ordinarily the injection of brain tissue from rabid animals into rabbits or guinea pigs incites the disease irregularly and after incubation periods of 2 to 8 weeks. Mice as test animals have proved in the past even less satisfactory.[1] Mice especially bred for

[1] Koch, J., *Kolle-Kraus-Uhlenhuth. Handb. der Path. Mikroorg.*, 1930, **8**, 547.

high susceptibility to neurotropic viruses, however, are more sensitive and uniform in their response to the rabies street virus.

The diagnostic test for rabies by mouse inoculation is carried out in the following manner: Fresh dog brain containing Negri bodies* is seared and dissected aseptically to expose Ammon's horn. This area is removed and portions emulsified, diluted approximately 10 times, centrifuged, and injected into mice intracerebrally in 0.03 cc. quantities and intraperitoneally in ½ cc. quantities. Some of the mice are sacrificed from the 5th to 8th days, their brains removed, and impression transfers made from Ammon's horn and stained to determine the presence of Negri bodies. The remainder are observed for the appearance of characteristic weakness and paralysis of hind legs, prostration, and death. Final tests for Negri bodies are made on sick and prostrate mice.

To date, 40 dog brains received by the New York City Department of Health Laboratories have been tested. In 32 specimens, no Negri bodies were found and the test mice remained well for 30 days. In 7 specimens, Negri bodies were demonstrable. Of these, 2 specimens, Nos. 1 and 3, were injected intracerebrally into 6 and 19 mice respectively. The 6 mice receiving specimen No. 1 remained well 10 days, were weak or paralyzed on the 11th day, and prostrate or dead on the 12th day. All showed abundant Negri bodies in stained impression transfers. Of the 19 mice given specimen No. 3, 2 were killed on the 5th and 2 on the 6th days and examined for Negri bodies. A positive diagnosis could not be made definitely on these mice. The remaining 15 mice became weak or paralyzed on the 10th to 13th days and all contained Negri bodies. The other 5 specimens, Nos. 2, 19, 29, 41 and 42, were injected both intracerebrally and intraperitoneally. Examinations for Negri bodies were questionable on the 5th day and positive on the 6th day. All remaining mice became weak or paralyzed on the 7th to 9th days and died on the 10th to 15th days. In these mice Negri bodies were invariably abundant. Brains of 28 supposedly normal skunks, squirrels, cats, rats, etc., have also been tested with negative results. Of the total 68 specimens, 2 contained bacterial contaminants fatal to the test mice. This brief experience suggests that by the intracerebral and intraperitoneal injection of special mice, rabies may be diagnosed within 7 days.

* The assistance of Dr. W. H. Park and Miss A. Mann, of the New York City Department of Health Laboratories, is gratefully acknowledged. Specimens from possible cases of rabies sent to the City Laboratory for diagnosis were preserved in clean condition at low temperature and after being studied, one-half of the whole brain was brought here for further testing.

TABLE I.

Protective Effect of Serum from Individual Receiving Semple Anti-Rabic Treatment Against Rabies Virus.

Serum	Virus serum dilutions. 0.03 cc. per mouse intracerebrally.							
	10^{-1}		10^{-2}		10^{-3}		10^{-4}	
	No. of mice injected	Duration of life in days	No. of mice injected	Duration of life in days	No. of mice injected	Duration of life in days	No. of mice injected	Duration of life in days
Broth control	3	8, 9, 10	3	9, 10, 10	3	11, 13, 13	4	14, 15, 15, 15
Untreated—W	4	9, 10, 10, 10	4	9, 9, 10, 10	4	12, 13, 15, 16	4	15, 16, 16, S*
Treated—D	4	13, 13, 13, 15	4	13, 13, 16, S	4	S, S, S, S	4	S, S, S, S

* S = Remained well 30 days.

The virus-containing dog brains proved infective for these mice in doses of 10^{-5} gm. when injected intracerebrally and 10^{-1} gm. when injected intraperitoneally or subcutaneously. Two to 5 intracerebral passages of the virus in these mice reduced the incubation period to 6 days without change in number, size, or appearance of the Negri bodies; the intraperitoneal and subcutaneous titres increased to 10^{-3}. The virus traverses Seitz filters in 10^{-1} and 10^{-2} dilutions when treated in the manner described by Bauer and Hughes.[2]

A mouse protection test for the quantitative measurement of protective antibodies against rabies virus is being developed. Brains from mice prostrate 8 to 9 days after intracerebral injection of mouse brain virus in the 2nd to 5th passage are emulsified, diluted, centrifuged, diluted again, combined with equal parts of test sera for 2 hours at 37°C. and 2 hours at 23°C., and then injected in 0.03 cc. quantities intracerebrally in mice in dilutions of 10^{-1} to 10^{-5}. The duration of life of the injected mice is recorded in days.

Five tests with serum from one individual 1½ years after receiving the last of 3 courses of Semple anti-rabic treatment and with sera from 4 untreated individuals have given uniform results. The protocol of the first test is summarized in Table I. Data thus far show that sera from the 4 untreated individuals do not protect mice against an intracerebral injection of 10^{-6} gm. of mouse brain virus of 4 different strains but that serum from the treated person does protect against at least 100 lethal doses of these same strains.

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Numerical Relations of an Unstable Variant of Salmonella Aertrycke.

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Although many bacteriologists have encountered variants which were difficult to stabilize, most have felt that with sufficient care and repeated selection, any variant could be obtained in a stable form. In our studies on colonial forms of *Salmonella aertrycke* certain variants were encountered which lacked stability. A variant of this

[2] Bauer, J., and Hughes, T. P., *J. Gen. Physiol.*, 1934, **18**, 143.