increased with the age of the embryo, as indicated by the increase in blackening on the x-ray film. These observations were supported by measurements with the Geiger-Muller counter; embryos of 8 to 12 days development gave no significant measurements. However, those of 13 days and older gave an increasing number of counts.

Microscopic study of sections of the thyroid of embryos from the 9th and 10th day of development revealed the presence of epithelial cells in irregular cords, but no discrete follicles. Thyroids of embryos of the 11th and 12th day contained a few groups of cells arranged in circular patches (young follicles), with a few discrete follicles in the 12-day embryos. There is a progressive increase in the number of follicles from the 12th through the 15th day of development with an increase in the quantity of colloid present.

Since it is assumed by many workers that the storage of colloid in the follicles indicates the function of the thyroid, then we may also assume that the initial appearance of colloid indicates the onset of thyroid function. If these assumptions are correct, then our observations support the conclusion that the thyroid of the embryo hamster begins to function on the 13th day of development, a short time after the first appearance of follicles.

It has been demonstrated for the embryos of many animals, including the human fetus (see introduction) that the storage of colloid is correlated with the differentiation of follicles in the gland, the quantity depending upon the degree of differentiation and the number of follicles present and these, in turn, are correlated with the age of the embryo. Our findings through autoradiographic and microscopic study support these conclusions for the thyroid gland of the golden hamster.

Summary. (1) Fifty to 100 microcuries of radioactive iodine in the form of NaI were injected into the peritoneal cavity of the gestating hamster on the 8th to the 15th day after copulation. (2) Observations made on embryos of from 8 to 15 days of development by means of autoradiographic study and tests with the Geiger-Muller counter show that radioactive iodine begins to accumulate in the thyroid on the 13th day of development. (3) The accumulation of radioactive iodine is coincident with the appearance of discrete follicles, which first appear between the 12th and 13th day of development. (4) The quantity of iodine-containing colloid increases with the number of follicles present and the age of the embryo.

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Cross-Immunity Tests in Animals with Epidemic Keratoconjunctivitis and St. Louis Encephalitis Viruses. (19112)

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Recent investigations disclosed that it is impossible to differentiate the viruses of epidemic keratoconjunctivitis (EK) and St. Louis encephalitis (SLE) by serological means(1,2). A hint that the two viruses may be different comes from the fact that EK caused a fatal encephalitis in guinea pigs and rabbits while SLE engendered only a non-fatal pyrexia in guinea pigs and nothing in rabbits. However, no distinct differences could be demonstrated by cross-immunity tests in animals. This report presents the data on the infectivity tests as well as the results of the cross-immunity tests.

Materials and methods. Viruses. The 2 strains of epidemic keratoconjunctivitis virus

1. Ruchman, I., PROC. SOC. EXP. BIOL. AND MED., 1951, v77, 120.
2. Cheever, F. S., PROC. SOC. EXP. BIOL. AND MED., 1951, v77, 125.

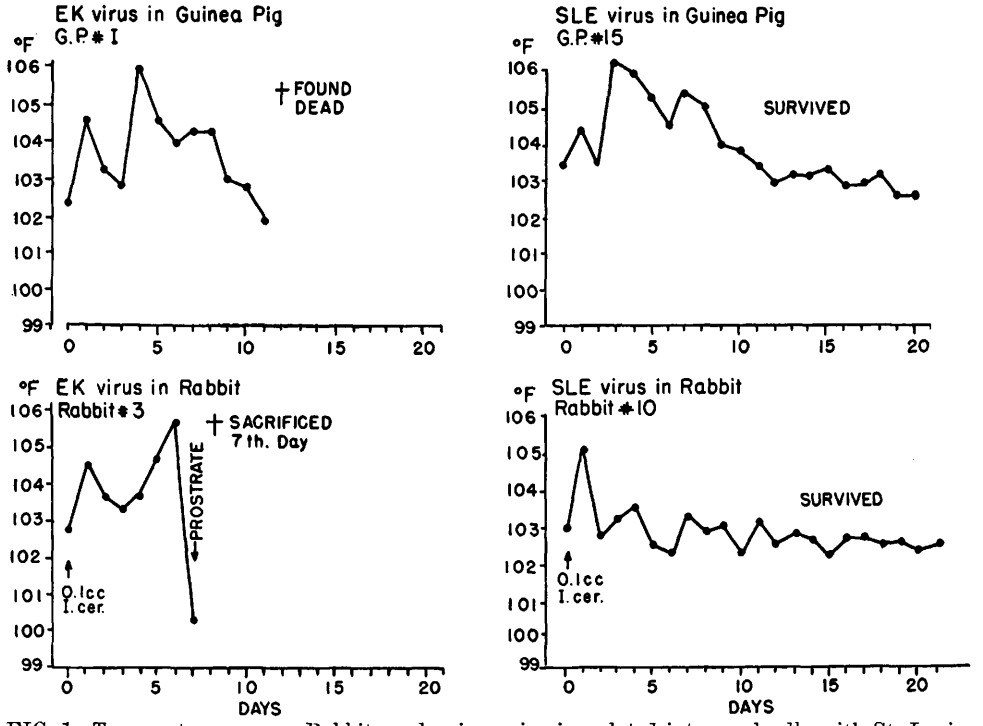

FIG. 1. Temperature curves. Rabbits and guinea pigs inoculated intracerebrally with St. Louis encephalitis and epidemic keratoconjunctivitis virus.

used in these tests were designated EK-103 and EK-TC17. They both came from Dr. A. E. Braley and were originally derived from the New York strain isolated by Dr. M. Sanders(3). Two strains of St. Louis encephalitis virus were used. One obtained from Dr. A. B. Sabin was designated *Webster* and the other supplied by Dr. W. McD. Hammon was designated *Hammon*. The Nakayama strain of Japanese B encephalitis virus (Jap B) was obtained from Dr. Sabin and the West Nile encephalitis virus (W. Nile) was supplied by Dr. Joel Warren. *Virus suspensions.* The virus suspensions were prepared by grinding infected mouse brains in a mortar with alundum and enough undiluted, normal rabbit serum to make a 20% suspension. After light centrifugation the supernatant fluid was distributed in 1 cc rubber stoppered vials and stored in dry ice. Virus dilutions were made in saline containing 10% normal rabbit serum. Although some variation occurred viruses used in this study generally titred 10^{-8} or higher. In a few instances 30% suspensions of EK infected rabbit brain were used to infect rabbits. *Animals.* The mice were albinos of either sex and were bred in our own laboratory. Those used in preparing the virus suspensions were between 18 and 24 days old and weighed approximately 8 to 11 g. Those that were used in the immunity studies were 6 to 8 weeks old and weighed around 20 g. All other animals were procured locally. The guinea pigs were of both sexes and weighed between 300 and 700 g. Rabbits also were of either sex and weighed between 2 and 3 kg. Albino rats were young males weighing approximately 150 g. Animals inoculated intracerebrally received 0.1 cc of a 10% suspension. Mice were only given 0.03 cc. The amount inoculated subcutaneously in mice was 0.1 cc of the designated dilution. Ether anesthesia was used for all intracerebral inoculations. As previously reported, the EK and SLE viruses behaved differently in rabbits and guinea pigs, and characteristic tem-

3. Sanders, M., *Arch. Ophthalmol.*, 1942, v28, 581.

TABLE I. Summary of Results in Animals.

Animals	Initial inoculation: Virus inoculated	Initial inoculation: No. tested	Result: Died	Result: Survived	Test for immunity: Virus inoculated	Test for immunity: No. tested	Test for immunity: No. immune
Guinea pigs	EK	12	10	2	SLE	2	2
	SLE	8	1*	7	EK	7	5†
Rabbits	EK	14	11	3	EK	1	1
	SLE	4	0	4	"	3	3
	Jap B	2	0	2	"	2	0
	West Nile	2	0	2	"	1	0
Rats	EK	6	0	6	—	—	—

* Died following cardiac puncture.
† Guinea pigs that died succumbed to intercurrent infection.

perature curves are shown in Fig. 1. Unless indicated otherwise the 103 strain of EK and the *Webster* strain of SLE were used in most of the tests.

Results. Several series of tests were performed and in most cases surviving controls were used for subsequent immunity tests. Results are shown in Table I. Twelve guinea pigs inoculated intracerebrally with the EK virus developed fever of several days' duration. Ten of them died between 7 and 28 days after injection. The 2 survivors were tested with SLE virus at a later date. They did not develop pyrexia and were therefore considered immune. Eight guinea pigs inoculated with SLE virus developed fever. One animal died following cardiac puncture 3 weeks after the initial inoculation and 7 survived. The 7 survivors were tested for cerebral resistance against the EK virus. Five survived and 2 died of an intercurrent infection. Thus animals immune to EK were also immune to SLE and conversely, animals resistant to SLE were also resistant to EK.

Fourteen rabbits injected intracerebrally with EK virus developed fever. Three inoculated with the TC17 strain died and 8 out of 11 inoculated with the 103 strain died. One of the 3 survivors was retested with the EK virus and it was found to be immune. It was impossible to test EK immune rabbits for immunity to SLE. Two rabbits each were inoculated intracerebrally with the *Webster* and *Hammon* strains of SLE. The animals remained afebrile. Three of the animals were tested with the EK virus at a subsequent date and they remained well. Two animals were available that had been hyperimmunized against Jap B and one was available that had been hyperimmunized against W. Nile. They had received numerous peripheral inoculations of living virus over a prolonged period of time. They were inoculated intracerebrally with EK and although they became febrile, they did not die. All 3 controls for this experiment developed fever but only one of them died. It thus becomes apparent that although rabbits immunized against SLE were resistant to the EK virus the same thing could not be demonstrated under the conditions of this experiment for rabbits immunized against Jap B and W. Nile viruses.

Three young adult white rats were inoculated intracerebrally with the TC17 strain of EK and 3 others were inoculated with the 103 strain. None of the animals showed any obvious signs of illness during the period of observation.

It was somewhat more difficult to get immune survivors among mice. Nevertheless some animals were obtained in the following manner. Twenty g mice were inoculated subcutaneously with 0.1 cc of a 10^{-2} dilution of virus. After a period of 2 to 3 weeks they were given 0.1 cc of a 10% suspension subcutaneously. Two weeks later the EK immunized mice and one month later the SLE immunized mice were inoculated intracerebrally with a 10^{-2} dilution of the homologous virus. After a waiting period of 5 weeks the survivors were tested for resistance against an intracerebral inoculation of a 10^{-2} dilution of the heterologous virus. The data appear in Table II. Seven mice that had survived a previous intracerebral injection of the EK virus also survived inoculation of the SLE

TABLE II. Intracerebral Cross-Immunity Tests in Mice.

Exp.	Tested with*	Previously immunized with†	Results in tested mice‡
1	SLE	EK	S,S,S,S,S,S,S
		Controls	3,3,3,4,4,4,4,5,5,5
2	EK	SLE	7,10,11,S,S,S
		Controls	2,3,4,4,4,4,4,4,5,5
3	Jap B	EK & SLE	4,4,7
		Controls	3,4,4,4,5,5,5,5
4	West Nile	EK & SLE	5,5,6,7
		Controls	4,4,4,5,5,5,6

* Test dose consisted of a 10^{-2} dilution prepared from an infected mouse brain suspension stored in solid carbon dioxide.

† Immunized mice had survived a previous intracer. challenge with a 1% suspension of the homologous strain of virus.

‡ Figure indicates day of death; S indicates survival.

virus. Conversely of 6 mice that were resistant to SLE, 3 were now resistant to the EK virus while 3 succumbed after prolonged incubation periods. Seven of the above survivors now immune to both EK and SLE were divided into 2 groups Three mice were examined for cerebral immunity to Jap B virus and 4 were tested against W. Nile virus. They all succumbed to a 10^{-2} dilution of virus inoculated intracerebrally.

Discussion. It was previously shown that by serological methods the viruses of epidemic keratoconjunctivitis and St. Louis encephalitis could not be distinguished(1,2). The present immunity results confirm the close relationship that exists between the 2 viruses and seemingly it is only by means of intracerebral inoculation in rabbits and guinea pigs that the viruses can be told apart. It is not apparent at the present time whether the results of the immunity tests in mice are of significance or whether they merely represent differences that might be found to occur among different strains of the same virus. The current results do not add anything new to previous reports but do confirm the remarkable similarities that exist between the viruses of epidemic keratoconjunctivitis and St. Louis encephalitis.

Summary. Intracerebral inoculation of epidemic keratoconjunctivitis virus caused a fatal encephalitis in rabbits and guinea pigs, whereas inoculation of St. Louis encephalitis virus caused a non-fatal pyrexia in guinea pigs and elicited no response in rabbits. Mice were equally susceptible to both viruses while rats were refractory. Guinea pigs recovered from EK infection were also immune to SLE and those animals recovered from SLE were immune to EK. Rabbits convalescent from SLE were immune to EK, but the reverse experiment could not be done. Although EK immune mice resisted SLE, the reverse was not completely true.

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Modification of Sensitivity to X Radiation by Morphine Sulfate.* (19113)

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Lowered oxygen tension has been shown to prevent damage from X radiation in a variety of organisms(1-4). Anoxia has been produced

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