sage transfers promptly at the time the febrile response occurred.

It may be of interest to mention here that other investigators[10,12-16] have used the concept of alternating passages to maintain, modify, reactivate or adapt viral or rickettsial agents to other hosts or tissues.

It is conceivable that it may not be essential to use the alternating passage method to pass hog cholera virus in rabbits since in work with rinderpest virus, Nakamura and colleagues[8] were able to adapt the virus to rabbits and maintain it for 166 consecutive serial passages without going back to the original host. Edwards[17] also reported success in maintaining rinderpest virus in rabbits for a period of at least 14 months by going directly from the calf to the rabbit and by making successive intravenous transfers in rabbits every 2-7 days. On the other hand Baker[1] found it necessary to use the alternating passage method to achieve the same object. The difference in results obtained by various investigators could be due to the fact that different breeds of test animals or different strains of virus may have been employed.

Summary. Hog cholera virus has been carried for 12 consecutive passages in rabbits by using infected rabbit spleen as transfer material. Starting from the 8th rabbit spleen passage, the virus was passed back to a pig for one passage and was then carried for 8 further passages in rabbits by using infected rabbit blood as transfer material. Aside from a febrile response, no other symptoms were observed in inoculated rabbits.

[12] Levaditi, C., Harvier, P., et Nicolau, S., *Annales de l'Institut Pasteur*, 1922, **36**, 107.

[13] Ch'en, W. K., PROC. SOC. EXP. BIOL. AND MED., 1933-34, **31**, 1252.

[14] Coffey, J. M., *Am. J. Pub. Health*, 1934, **24**, 473.

[15] Cox, H. R., *Science*, 1941, **94**, 399.

[16] Waddell, M. B., and Taylor, R. M., *Am. J. Trop. Med.*, 1945, **25**, 225.

[17] Edwards, J. T., Report of the Imperial Bacteriological Laboratory, Muktesar, India, for the 2 years ending March 1924, p. 32.

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Serial Passage of Hog Cholera Virus in Rabbits.

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Successful adaptation of rinderpest virus of cattle to the rabbit[1] naturally led to similar experiments with hog cholera virus since both of these agents are quite specific for their natural hosts. In the original work on rinderpest alternate transfer of virus from its natural host to the rabbit and then back to the calf was the procedure that made for success. With this alternating technic virulence of the rinderpest virus for the rabbit increased progressively and, following this increase, evidence of disease was found in the rabbit. The results of the experiments on hog cholera virus follow.

Virus Strains Used. Strain A was obtained through the courtesy of Dr. TenBroeck[2] and, as reported by him, it was one used for vaccination in the central portion of the United States. In his work this strain was grown in the presence of fresh minced swine testicle on the chorioallantoic membrane of embryonated eggs for 13 transfers, followed by an equal number of transfers on agar, making 26 transfers in all. From the last transfer lyophilized material was kept from April 5, 1941 until March 24, 1945 when it was injected into a pig. For further preservation spleen from this animal was kept frozen with

[1] Baker, J. A., *Am. J. Vet. Research*, 1946, **7**, 179.

[2] TenBroeck, C., *J. Exp. Med.*, 1941, **74**, 427.

TABLE I.
Effect on Swine of Hog Cholera Virus (Strain A) Before and After Serial Passage in the Rabbit.

No. of serial transfers in rabbits	Swine		Results in swine				
	No.	Wt, lb	Incubation period, days	Duration of fever, days	Highest temp., °C	Outcome of illness	Reaction to injection of virulent virus
0	1	37	3	9	41.0	Died	
0	22	27	3	11	41.5	"	
0	29	30	3	10	41.3	"	
0	37	24	3	11	41.8	"	
0	40	21	3	8	41.5	"	
5 (a)*	8	80	3		41.5	Killed	
5 (b)	11	70	3		41.2	"	
10 (b)	13	70	?	0	39.3	Survived	Immune
10 (b)	20	25	?	0	39.7	"	"
10 (a)	27	30	3	4	41.3	"	"
15 (b)	42	32	5	2	40.6	"	"
15 (a)	43	30	4	3	40.6	"	"

* (a) indicates first group of serial passages; (b) second group of serial passages.

dry ice until January 2, 1946 when this work began.

Strain B was obtained through the courtesy of Dr. Shope. This strain was recovered by him from a natural outbreak May 26, 1944 and was subsequently carried through 5 transfers in swine. In interim periods between transfers the virus was kept as infected blood in a refrigerator.

Rabbits Used. The rabbits used were from mongrel stock, largely Polish-Dutch, from 3 to 5 months old weighing 1500 to 2500 g.

Procedure Used. Each of 2 rabbits was inoculated intravenously with 1 cc of a 10% suspension of spleen obtained from an infected pig at the height of illness. The rabbits were killed after 5 days and a portion of spleen from each was used for making another 10% suspension. Two rabbits were given 1 cc each of this suspension and injections of the same suspension were made into swine to determine the amount of virus present. Spleen tissue was also frozen in CO_2 and was available for retest if necessary. Tests for contaminating organisms made on blood agar slants and sealed meat medium showed occasional cocci from a few of the pig spleens. Suspensions of rabbit spleens showed no growth. Temperatures were taken and observations made on the rabbits and swine in the morning and evening. Swine that died were autopsied and the diagnosis was made on the finding of typical lesions. Any animals that survived were reinoculated with at least 100 times the infective dose and a control animal injected with the same virus was kept in the same room until its death. If the surviving animal showed no signs of illness from this test injection and exposure period, it was evident that hog cholera virus had been present in the material used for the first inoculation.

Results. Strain A. Test of the spleen suspension of the first rabbit passage showed that 0.01 g of tissue would infect swine, whereas 0.001 g would not. The spleen suspension of the 2nd and subsequent rabbit passages was not titrated, but 0.1 g contained virus, as shown by swine inoculation on the 2nd, 5th, 10th, and 15th passages. These results were repeated in a similar series using as starting material spleen from a different pig and the composite results are shown in Table I.

As Table I shows, strain A was twice passed serially through rabbits for 15 transfers. There was noted a decrease in virulence for swine, as rabbit spleen suspensions from the 10th and 15th serial passages injected intramuscularly produced fever of short duration as the only sign of illness (Table I and Fig. 1). This was followed by complete immunity to an intramuscular injection of 100 times the amount of fully virulent hog cholera virus that produced death in a control animal kept in the same room. Virus attenuated in this way by serial passage in rabbits may make a practical and inexpensive vaccine.

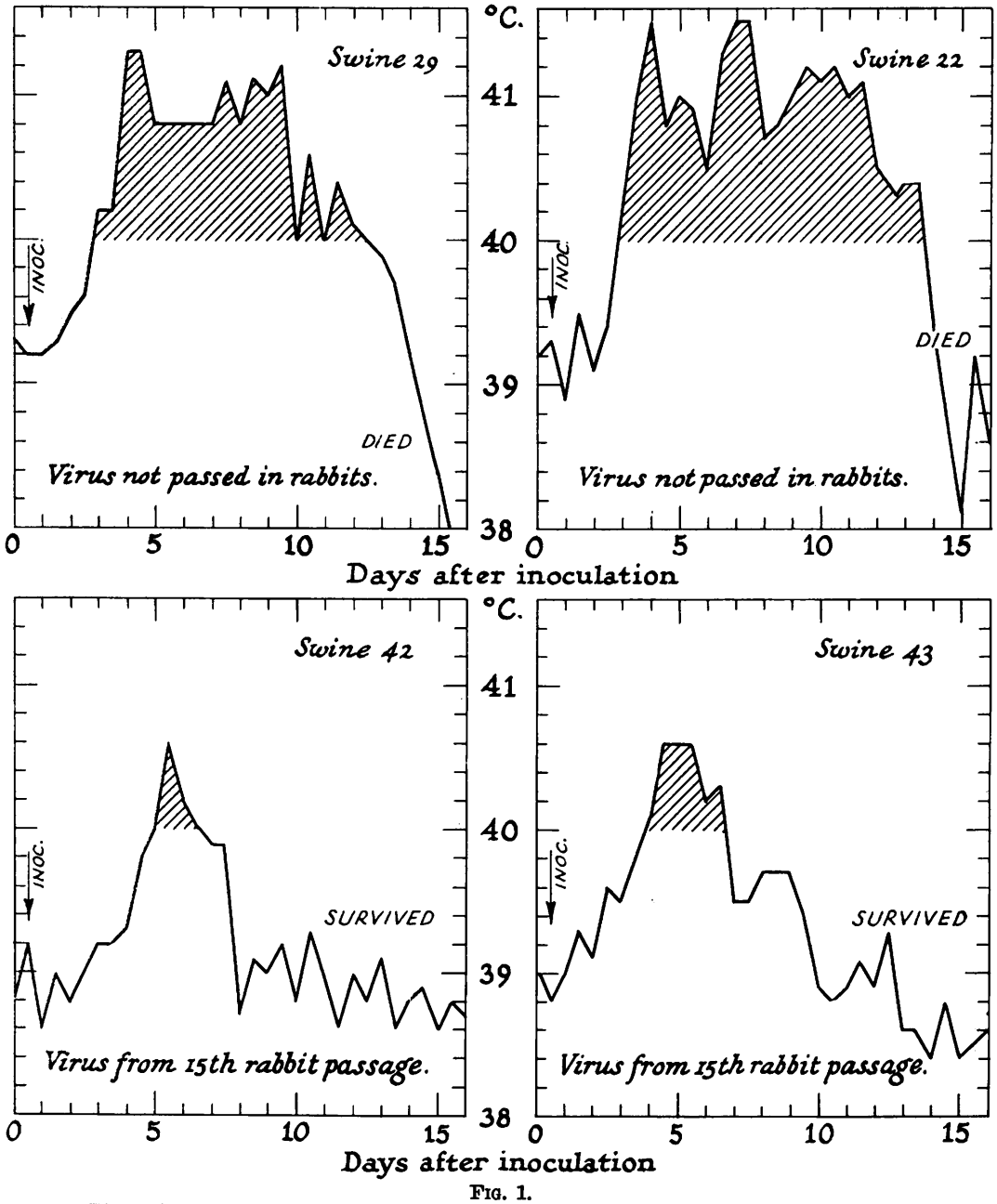

FIG. 1.

Thermal reaction in pigs given hog cholera virus (strain A) not transferred in rabbits and from the 15th serial passage.

Strain B. In the first passage 0.1 g of the rabbit spleen infected pigs but 0.01 g did not. Transfer to a second rabbit failed, since 0.1 g of this spleen did not infect a pig. The alternating technic described briefly above and more fully in the rinderpest paper[1] was then attempted. As Table II shows, swine were infected with 0.1 but not with 0.01 g of spleen from rabbits in the first 3 alternating passages. In no case was the virus detected in the 2nd consecutive rabbit passage. After the 4th and 5th alternation, 0.01 g of

TABLE II.
Hog Cholera Virus (Strain B) Alternated Between Swine and Rabbits.

Alternating passage in swine	Serial passage in rabbits	Test of rabbit spleen for virus by swine injection	
		Amt (g)	Result
1st	1	0.1	+*
		0.01	—
	2	0.1	—
2nd	1	0.1	+
		0.01	—
	2	0.1	—
3rd	1	0.1	+
		0.01	—
	2	0.1	—
4th	1	0.01	+
		0.001	—
	2	0.1	—
5th	1	0.01	+
		0.001	—
	2	0.1	—
6th	1	0.001	+
		0.0001	—
	2	0.1	+
	3	0.1	—
7th	1	0.001	+
		0.0001	—
	2	†	
	3	0.1	—
8th	1	0.001	+
		0.0001	—
	2	†	
	3	0.1	—

* + indicates virus present; —, no virus.
† Not tested.

spleen then infected swine. Again no virus was detected in the 2nd consecutive rabbit passage. After the 6th, 7th, and 8th alternation, 0.001 g then infected swine. With this last increase of virus in the rabbit, transfer was successful for 2 but not 3 serial rabbit passages.

Swine inoculated with infective spleen suspensions from rabbits in the 1st, 2nd, and 3rd alternating passages showed an incubation period of 3 days while a time interval of 4 to 7 days between injections and appearance of fever was noted in those given a similar amount of virus from subsequent alternating passages. It seems evident, therefore, that adaptation of strain B for rabbits was caused by alternating the virus between swine and rabbits.

Effect of Hog Cholera Virus on Rabbits. None of the rabbits inoculated with either strain A or B showed any temperature elevation in early passages. In later passages a slightly increased temperature was noted in some animals inoculated with either strain but temperatures exceeding 40°C seldom occurred. No other signs of illness were noted. None of the animals upon autopsy showed lesions referable to hog cholera.

Discussion. A review by Muir[3] of the divergent results obtained by others in transfers of hog cholera virus to animals other than swine would suggest that some strains are adaptable while others are not. The complete adaptation to rabbits of strain A

[3] Muir, R. O., *J. Comp. Path. and Therap.*, 1940-1943, **53**, 237.

and incomplete adaptation of strain B in our work would support this idea. Also, adaptation of yellow fever by Theiler and Smith,[4] dengue by Sabin and Schlesinger,[5] and perhaps other viruses to animals has not been obtained regularly.

The fact that strain A had been kept under laboratory conditions for a long period and had been cultured outside the body of swine may have influenced its adaptability to the rabbit, although it is possible that strain A was naturally more adaptable. On the other hand, strain B had been isolated recently. It showed poor adaptability and required 6 alternating passages before virus increase occurred in the rabbit spleens and it could be transferred consecutively in the rabbit.

Since neither strain A nor B produces definite signs of illness in rabbits, a study of other strains and perhaps in other laboratory animals should be made especially by the alternating method which, as was shown with strain B and rinderpest, allows opportunity for development of any latent pathogenic potentiality. Such a study may give results similar to those obtained with rinderpest and supply the laboratory animal that has been needed in hog cholera research.

Summary. Two strains of hog cholera virus were inoculated into rabbits. One strain (A) continued in serial passage, became attenuated for swine and, after 15 rabbit passages, produced a febrile reaction of short duration as the only sign of illness. This attenuated strain fully immunized swine to the virulent hog cholera virus. The other strain (B) was present in rabbits for 1 but not 2 consecutive passages. With 6 continued transfers alternately between swine and rabbits, the amount of virus in the rabbit's spleen attained 100 times that found in the initial rabbit transfer. Serial passage in the rabbit was then successful for 2 but not 3 transfers. Evidence of attenuation of this strain was the lengthened incubation period in swine inoculated with the rabbit-passed material. Possible explanations for the difference in adaptability of these strains are discussed.

[4] Theiler, M., and Smith, H. H., *J. Exp. Med.*, 1937, **65**, 767.

[5] Sabin, A. B., and Schlesinger, R. W., *Science*, 1945, **101**, 640.

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Antihistaminic Substances in Histamine Poisoning and Anaphylaxis of Mice.

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The conditions under which mice can be sensitized and the manifestations of sensitization in mice closely simulate those of anaphylaxis in guinea pigs and other animals (Braun,[1] Schultz and Jordan,[2] Ritz,[3] Sarnowski,[4] Bourden,[5] Weiser, Golub and Hamre[6]). Active sensitizations can be produced with various protein substances, horse, sheep, cow, guinea pig serum, or egg white; passive sensitizations with immune rabbit serum, antihorse guinea pig serum, antihorse rabbit serum, antipneumococcus Type I rabbit serum, etc. The sensitizations are specific and their duration varies from several weeks to several months. Refractoriness occurs after recovery from shock and active desensitization is obtained by

[1] Braun, H., *Münch. med. Wschr.*, 1909, **37**, 1880; *Z. Immunf.*, 1910, **4**, 590.

[2] Schultz, W. H., and Jordan, H. E., *J. Pharm. and Exp. Therap.*, 1911, **2**, 375.

[3] Ritz, H., *Z. Immunf.*, 1911, **9**, 321.

[4] Sarnowski, V., *Z. Immunf.*, 1913, **17**, 577.

[5] Bourdon, K. L., PROC. SOC. EXP. BIOL. AND MED., 1937, **36**, 340.

[6] Weiser, R. S., Golub, O. J., and Hamre, D. M., *J. Inf. Dis.*, 1941, **68**, 97.