and acetoacetic acid. 3. The synthesis increased in the presence of butyric-, propionic-, valeric-, and *n*-caproic acid. 4. The synthesis decreased in the presence of formic-, *n*-caprylic-, capric-, lauric-, oleic-, linoleic acid, glyceraldehyde, acetylaldehyde, and β-hydroxy butyric acid.

15046

Modification of Serological Response to Infection with Murine Typhus by Previous Immunization with Epidemic Typhus Vaccine.

HARRY PLOTZ* AND KENNETH WERTMAN.

From the Division of Virus and Rickettsial Diseases, Army Medical School, Washington, D.C.

It is well known that epidemic typhus cannot be differentiated from murine typhus by means of the non-specific Weil-Felix test since a high OX-19 agglutination titer may occur in both diseases. However, within the past few years, through improved technics and experimentation, both the specific complement fixation and agglutination tests have become of great value in differentiating these diseases. As was first shown in this laboratory, there is an antigenic fraction (soluble antigen) common to both types of rickettsiæ which reacts equally well in the complement fixation test with epidemic or murine antibodies.[1,2,3] However, by washing and centrifugation this antigen can be eliminated, leaving the purified rickettsial bodies. These latter constitute a highly specific antigen in the complement fixation test. Furthermore, these rickettsial suspensions have been found to be of great value as agglutinogen in the agglutination test. With technical improvements, recently described, this test has also been found to be of considerable value as a means of differentiating epidemic from murine infections.[4] As illustrations of the type of serological reactions found see Tables I and II which are representative cases selected from our series where the agent was isolated and identified and where serial serum specimens were available for examination.

It is noted that in epidemic typhus high complement-fixing titers are obtained with the purified epidemic rickettsial antigen and low or negative results with a purified murine rickettsial antigen. Conversely, in murine typhus high titers are obtained with a purified murine rickettsial antigen and low or negative results with a purified epidemic antigen. With regard to the rickettsial agglutination results higher titers are obtained with the homologous antigen than with the heterologous. However, it is to be noted that while there is cross agglutination the difference in titers is sufficient to permit clear differentiation.

During the past year we have received specimens from 147 cases of murine typhus occurring in soldiers stationed in the United States. Of these 135 gave a typical murine serological response comparable to that cited in Table II. However, 12 cases have given an unusual response. Table III records the serological results found in these cases.

In an attempt to determine the factors producing these atypical responses the 12 cases

* Member of the U. S. A. Typhus Commission.

1 Report on the Testing of Four Different Typhus Vaccines in Guinea Pigs. Report to the Director, Army Medical School, 20 March, 1942, to be published.

2 Plotz, H., *Science*, 1943, **97**, 20.

3 Plotz, H., Wertman, K., and Bennett, Report to the Director, U. S. A. Typhus Commission, 15 February, 1944, to be published.

4 Plotz, H., and Snyder, Report to the Director, U. S. A. Typhus Commission, 31 October, 1944, to be published.

TABLE I.
Serologic Results in a Non-vaccinated Individual Infected with Epidemic Typhus Fever. Case 2724—Strain Isolated.

Day of disease	Weil-Felix*			Comp.-Fixation†		Rickettsial Aggl.	
	OX-19	OX-2	OX-K	Epidemic	Murine	Epidemic	Murine
8	1/20	0	0	1/40	0	1/80	0
11	1/320	1/40	0	0	0	1/160	1/40
13	1/2560	1/80	0	1/20	0	1/1280	1/320
15	1/2560	1/80	1/20	1/80	1/10	No serum	
19	1/2560	1/80	0	1/640	0	1/5120	1/1280
23	1/5120	1/80	0	1/2560	0	1/10,240	1/1280
25	1/2560	1/40	0	1/1280	0	1/5120	1/1280
27	1/1280	1/40	0	1/1280	0	1/5120	1/640
86	1/640	1/40	0	1/160	0	1/640	1/40
286	1/80	1/10	0	1/160	0	1/80	1/80

* Weil-Felix and rickettsial agglutination titers are recorded as final dilution.
† Complement fixation is recorded as initial dilution.

TABLE II.
Serologic Results in a Non-vaccinated Individual Infected with Murine Typhus Fever. Case I.—Strain Isolated.

Day of disease	Weil-Felix			Comp.-Fixation		Rickettsial Aggl.	
	OX-19	OX-2	OX-K	Epidemic	Murine	Epidemic	Murine
5	0	0	0	0	0	0	0
7	1/80	0	0	0	0	0	1/160
9	1/640	1/160	1/40	0	0	1/80	1/1280
11	1/640	1/160	1/80	0	0	1/160	1/2560
13	1/640	1/160	1/40	0	0	1/320	1/5120
14	1/640	1/80	1/40	0	0	1/320	1/5120
16	1/640	1/80	0	0	0	1/320	1/5120
18	1/320	0	0	0	1/80	1/320	1/5120
20	1/160	0	0	0	1/160	1/640	1/10,240
22	1/160	0	0	0	1/320	1/640	1/10,240
31	1/160	0	0	0	1/320	1/320	1/5120
75	0	0	0	1/40	1/320	1/40	1/1280
123	0	0	0	1/10	1/320	0	1/160

were investigated with some care. It was found that all of these patients had received typhus vaccine some time or other within the preceding 2 years. The vaccine used consisted of formalinized epidemic rickettsiæ with no murine rickettsiæ whatsoever. All cases occurred in regions of the United States where murine typhus alone is known to exist and in some instances other cases of murine typhus were present in the same camp at the time these cases were admitted to the hospital. The clinical diagnosis of murine typhus seems to have been well substantiated. All cases developed a typical rash. A rise in OX-19 titers occurred in the late febrile period and early convalescence in 6 cases, while most of the others showed high titers. The febrile period varied from 8 to 23 days with an average of about 14 days. Unfortunately, no attempt was made to isolate the agents.

From the figures given in Table III it is apparent that there is frequently a reversal between the results of the complement fixation tests and the agglutination tests and even the quantitative relationships are dissimilar to those previously found in individuals who had never received vaccine. It is observed that while many of these specimens showed higher epidemic complement fixation titers than was found with the murine antigen there was usually considerable cross fixation and a number of specimens showed equal titers to the 2 antigens. This is in contrast to what was found in non-vaccinated individuals infected with epidemic typhus where high epidemic titers are obtained and where con-

TABLE III.

Serologic Results Found in Individuals Who Were Previously Immunized with Epidemic Typhus Vaccine and Who Subsequently Contracted Murine Typhus.

Case	Day of disease	Typhus vac.	Compl.-Fix. Epi.	Compl.-Fix. Mur.	Rick. Agg. Epi.	Rick. Agg. Mur.	Weil-Felix* OX-19	Weil-Felix* OX-2	Date of onset	Where infected	Severity of infection
A	59	11/43	1280	320	80	640	80	80	1/1/44	Waycross, Ga.	Moderately severe
	90		1280	320	160	1280	0	0			
S	21	9/42	640	320	80	1280	2560	0	11/20/44	Ft. Benning, Ga.	" "
	42		320	320	80	640	640	0			
	58		160	160	80	320	640	0			
Si	27	5/27/44	A/C	A/C	2560	10240	160	40	7/19/44	Louisiana	Severe
	159		320	160	320	2560	160	40			
P	22	9/44	1280	640	320	640	160	0	12/17/44	Camp Gordon, Ga.	"
	50		1280	640	320	320	0	0			
B	18	11/44	160	40	5120	10240	640	0	11/23/44	Camp Shanks, N.Y.	Moderately severe
	21		160	160	5120	10240	320	0			
	26		160	160	5120	10240	0	0			
	31		160	160	5120	10240	0	0			
Be	18	8/44	640	160	5120	10240	1280	0	11/25/44	Birmingham, Ala.	" "
	70		320	160	160	1280	160	0			
Ben	38	10/44	80	80	1280	5120	640	40	10/24/44	Tampa, Fla.	Mild
K	30	9/44	160	80	5120	10240	80	0	9/18/44	Camp Miles Standish, Mass.	"
Ba	15	12/44	80	10	1280	5120	640	320	12/15/44	Robins Field, Ga.	"
F	44	1/44	80	80	640	5120	640	0	1/2/45	Austin, Texas	Moderately severe
	55		80	40	160	2560	1280	40			
T	45	7/44	1280	640	1280	5120	1280	160	1/14/45	San Antonio, Texas	Mild
Com	14	8/43	320	320	160	640	160	0	7/12/44	Hunter Field, Ga.	"
	23	Recall dose	A/C	A/C	1280	2560	160	40			
	24	5/44	320	320	1280	5120	80	40			
	30		320	160	320	2560	40	0			
	37		320	160	320	2560	0	0			

* Cases S, P, B, Ba, and T showed a rise in OX-19 titer when serum specimens were examined during the febrile period and early convalescence while the other cases showed high titers. These examinations were made in the hospitals where the patients were treated. It should be observed that the specimens which were available for this study were frequently obtained late in convalescence.

siderable cross fixation is not the rule. Similarly, the complement fixation results are not comparable with what is found in non-vaccinated individuals infected with murine typhus, for here again the homologous complement fixation titer is higher than the heterologous with little crossing (see Table II).

In contrast to the complement fixation results, the rickettsial agglutination usually showed a higher murine titer. However, as in the complement fixation results cross agglutination is marked and in many instances there is only a 2-fold difference in titer. The same type antibody response was observed in the cases reported irrespective of whether the epidemic typhus vaccine was administered two years or one week before the present attack of murine typhus. Insofar as no such reaction has been observed in vaccinated subjects who subsequently became infected with atypical pneumonia, measles, infectious hepatitis, smallpox, pneumonia, meningitis, or other virus, bacterial, protozoal infections, it is our belief that the unusual serological findings reported are to be attributed to the previous immunization with rickettsial products followed by an infection with murine rickettsiæ.

It is of interest to observe, as far as can be judged from the description of the symptoms and course of the disease, that the previous administration of the epidemic typhus vaccine had little or no effect upon the severity of the recent attack of murine typhus.

Summary. Twelve cases of murine typhus occurring in individuals who previously received epidemic typhus vaccine are reported. It appears that the previous administration of the epidemic typhus vaccine did not affect the course or severity of the murine disease. The serologic response was different from that found in non-vaccinated cases of epidemic or murine typhus. The Weil-Felix reaction does not seem to be affected, for a rise in titer or high titers were obtained in a majority of the cases. The difference occurred in the specific antibody response. While the epidemic complement fixation titers were higher than the murine titers, a number of specimens showed equal titers or considerable cross fixation. The murine agglutination titers were usually higher than the epidemic titer with considerable cross agglutination. This type of reaction has not been found in individuals who had received epidemic typhus vaccine and who subsequently became infected with bacterial, viral, or protozoal infections. Since many of our soldiers who have received epidemic typhus vaccine may subsequently develop murine typhus either abroad or after return to this country, it is obvious that we may expect to have a considerable number of future cases presenting the serological pattern described in this paper.

15047

Use of Vaginal Smears in Mating the Golden Hamster.

GEORGE C. KENT, JR., AND JOHN P. MIXNER. (Introduced by Wm. H. Gates.)
*From the Department of Zoology, Louisiana State University, Baton Rouge.**

Various procedures have been adopted in different laboratories for breeding the golden hamster, *Cricetus* (*Mesocricetus*) *auratus* Waterhouse. The general procedure involves placing the male with one or more females for varying lengths of time, then removing the male and watching for signs of pregnancy. The male may be housed with the female during 2 or 3 estrous cycles,[1] or for 7 or 8 days,[2] or the male may be placed in the cage with several females after 6:30 p.m. and, if mating activity is observed, the reacting animals are left together over night.[3] If no mating activity

* Contribution No. 73.

[1] Peczenik, O., *J. Endocrin.*, 1942, **3**, 157.

[2] Sheehan, J. F., and Bruner, J. A., *Turtox News*, 1945, **23**, 65.

[3] Bond, C. R., *Physiol. Zool.*, 1945, **18**, 52.