than the cortex of the domestic rat, it is apparent that the wild rat has much more cholesterol available for hormone production than does the domestic rat of comparable size. The writer is unable to offer any explanation for the disparity in the two sets of figures by different investigators shown in the table.

It should be pointed out that these rats were caught in spring steel traps and in some cases may have remained in the trap for as long as 12 hours. The emotional disturbances and trauma associated with this may have caused a lowering of the original cholesterol.

Summary. Quantitative analysis of the adrenal glands of the wild Norway rat reveals that the total fats comprise 6.5% of the gland and that of this 3.7% are cholesterols. These figures are lower than the values for the adrenal gland of the domestic rat. However, since the cortex of the adrenal gland of the wild rat is 2-3 times larger than that of the domestic rat, the wild rat has much more cholesterol available for hormone production.

16608

Complement Fixation in Human Sera Following Murine Typhus.

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Complement-fixing antibodies for murine typhus have been shown to persist in the sera of both human beings and rodents for long periods of time following infection.[1-3] The complement fixation test has been employed, therefore, to estimate the past incidence of typhus in human[4] and rodent[5] populations. For this reason, an appraisal of the accuracy of the test as an indicator of past infection should be of interest. During a recent state-wide survey of murine typhus in Florida,[6] clinical and epidemiological records were completed on 2055 persons who, during the years 1944, 1945 and 1946, had had typhus or had been strongly suspected of having had the disease. The present report will describe the results of complement fixation tests performed upon the sera of 404 persons whose cases were investigated during the survey.

Methods. All persons from whom serum specimens were obtained had had fever and constitutional symptons sufficiently severe to confine them to bed for a period of at least 10 days. Furthermore, all these persons had had relatively close contact with rats and their ectoparasites, and there was epidemiological evidence that they could have contracted typhus.

Sera were collected aseptically at intervals varying from 7 days to 3 years and 11 months following the onset of illness, and were stored at icebox temperature for periods varying from 7 days to 4 months before examination. Immediately before testing they were inactivated by exposure to a temperature of 56°C for 30 minutes.

Antigen* was prepared by the Cox method[7] from the yolk sacs of chick embryos infected

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2 Bengtson, I. A., and Topping, N. H., *Am. J. Pub. Health*, 1942, **32**, 48.

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4 Davis, D. E., and Pollard, M., *Pub. Health Rep.*, 1946, **61**, 928.

5 Davis, D. E., and Pollard, M., *Am. J. Trop. Med.*, 1946, **26**, 619.

6 Rickard, E. R., and Riley, E. G., *Am. J. Pub. Health*, to be published.

* We are grateful to Dr. Herald R. Cox of the Lederle Laboratories for furnishing the antigens used in this study.

7 Cox, H. R., *Pub. Health Rep.*, 1938, **53**, 2241.

with murine typhus rickettsiae of the Wilmington strain. Lipoids were extracted with benzol and ether, and the antigen was purified by sodium sulfate precipitation in order to eliminate or reduce non-specific reactions with syphilitic sera.[8]

Hemolysin was titrated in 0.25 ml amounts in twofold dilutions, with starting dilutions of 1:200 and 1:300 followed by the addition of 0.25 ml of 3% washed sheep cells and 0.5 ml of guinea pig complement diluted 1:30. The highest dilution of amboceptor which showed complete hemolysis after incubation for 30 minutes at 37°C constituted one unit. This dilution divided by 3 was employed in the tests. Complement was titrated in 0.5 ml amounts in the presence of 0.25 ml of diluted antigen and 0.25 ml of normal saline. The hemolytic system of 0.5 ml of 1.5% sensitized cells was added, and results were read after incubation for 30 minutes at 37°C. Two full units of complement in 0.5 ml amounts were used in the tests. Antigen was titrated in serial twofold dilutions of 0.25 ml amounts with serial twofold dilutions of the same amounts of a positive serum. Complement was added and, after 18 hours of icebox fixation, the above-mentioned hemolytic system was added. The titration was read after incubation for 30 minutes at 38°C. The highest dilution of antigen giving 4 plus fixation with the highest dilution of serum was considered the unit of antigen. Two units in 0.25 ml amounts were used in the tests.

Serum specimens were diluted in 0.25 ml amounts in original dilutions ranging from 1:2 to 1:128. Tests were carried out in the same manner as described under titration of antigen, with the exception that the highest serum dilution in which 3 plus or greater fixation was observed was considered the end point. In tests for each day, 2 known positives and 3 known negative control sera were included, as well as a control titration of the complement with the same amount of antigen as was used in the test. Anticomplementary controls were included for all sera in the lowest dilution, and anticomplementary sera were

[8] Van der Scheer, J., Bohnel, E., and Cox, H. R., *J. Immunology*, 1947, **56**, 365.

TABLE I.
Correlation of Clinical and Weil-Felix Agglutination Findings During Illness with Titers Observed in Late Complement Fixation Tests.

Group No.	Findings at time of illness	No. sera examined	No. positive sera	% positive	Titers Complement fixation tests 0	1:2	1:4	1:8	1:16	1:32	1:64	1:128
I	Rash. Weil-Felix 1:320 or more	124	109	88	15	2	5	18	34	27	19	4
II	No rash. Weil-Felix 1:320 or more	79	67	85	12	1	6	13	14	22	7	4
III	Rash. No serological exam.	105	77	73	28	0	5	14	12	25	14	7
IV	Rash. Weil-Felix 1:160 only	21	14	67	7	0	1	3	4	5	1	0
V	No rash. Weil-Felix 1:160 only	16	8	50	8	0	0	2	3	3	0	0
VI	No rash. No serological exam.	59	38	65	21	2	3	2	15	10	4	2
		—	—	—	—	—	—	—	—	—	—	—
	Total	404	313	77	91	5	20	52	82	92	45	17

TABLE II.
Titers of Positive Complement Fixation Tests in Relation to Time After Onset of Illness.

Months after onset of illness	No. positive sera	Titers							Mean positive titers
		1:2	1:4	1:8	1:16	1:32	1:64	1:128	
12	29	0	2	3	2	11	5	6	1:34
12-23	67	0	1	6	17	24	12	7	1:30
24-35	123	4	6	28	31	38	16	0	1:18
36-47	94	1	11	15	32	19	12	4	1:18
Total	313	5	20	52	82	92	45	17	1:21

excluded from the series.

Results. In Table I, the persons on whose sera complement fixation tests were performed have been classified in groups according to the history of the presence or absence of a typical typhus rash and the presence of Weil-Felix agglutination in significantly positive or suggestive titer at the time of their illnesses. It would seem highly probable that the persons in Groups I and II had suffered from murine typhus. Weil-Felix agglutination titers of 1:320 or more are generally considered as diagnostic of rickettsial infection. Rocky Mountain spotted fever was the only rickettsial disease known to occur in Florida which might have caused the symptoms of, and the findings on, the patients in these two groups. In no instance did epidemiological evidence suggest this disease. Moreover, of the 91 sera negative for typhus, 66 were examined by the same complement fixation test for Rocky Mountain spotted fever. With the exception of 13 sera in which positive results were obtained in 1:2 dilution only, all of the sera examined for Rocky Mountain spotted fever were negative. Because of the lack of both clinical and epidemiological evidence of Rocky Mountain spotted fever, these positive results in low titer were considered non-specific.

From the data on Groups I and II presented in Table I, it would appear that the complement fixation test was approximately 85 to 88% accurate as an indicator of past infection with murine typhus for periods up to 3 or 4 years following illness. It was also apparent that positive titers were quite high. The most common titer was 1:32, and the mean was 1:21. In the individuals included in Groups III to VI, the diagnosis of typhus was not considered completely established in every case. The possible inclusion of diseases other than typhus in these groups might account for the lower percentage of positive sera observed in the groups. This fact appeared to have no influence on the mean positive titers for Groups I and II as compared with Groups III to VI with titers of 1:21 and 1:22 respectively.

In Table II, persons with positive titers have been classified in one-year periods according to the time elapsed between illness and the taking of serum for complement fixation. The diminution in mean positive titers observed in the yearly intervals after infection was not statistically significant. This observation suggested that no pronounced loss of positive titer had occurred with the passage of time.

As a control upon the specificity of the test, the sera of 20 inmates of a New York State correctional institution were examined for murine typhus by the same technic. Of these sera, 17 were negative and 3 fixed complement in 1:2 dilution, one serum with 4 plus fixation and 2 with 3 plus. None of the 3 subjects had positive serological reaction for syphilis. Two had never been out of New York State and had never taken typhus vaccine. One, however, had had 2 years of naval service on the island of Guam ending a few months before the specimen was taken. It was, therefore, possible that this person had received typhus vaccine or may have been infected during the course of his travels. However, there had been no history of illness during the period of his absence from New York State. It would appear that at least 2, and probably all 3, of these positive reactions were nonspecific. In consideration of the higher titers commonly observed in persons who had had typhus, the occasional oc-

currence of non-specific reaction in low dilution of serum would not seem to detract greatly from the utility of the test.

Summary. Complement fixation tests for murine typhus were performed upon the sera of 203 persons who, it is highly probable, had suffered an attack of murine typhus 7 days to 3 years and 11 months previously. The tests were positive in 85 to 88% of the cases, and in most instances the positive titers were high. Among 201 persons suspected of having had typhus, though the diagnosis was not conclusively established, the per cent of positive reactions was less, but the titers of positive sera did not differ significantly from those of the group in which the diagnosis was considered to be established. Up to periods of 4 years after infection, mean positive titers at yearly intervals did not suggest a pronounced loss of titer with passage of time. With the exception of infrequent reactions at 1:2 dilutions in the sera of persons who had not had typhus, the test was found to be specific.

16609

Studies on Phosphorus-Containing Compounds in Normal and Polioencephalitic Mouse Brain.*

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(Introduced by M. B. Visscher.)
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Chemical investigations in the field of neurophysiology have shown that all conditions leading to a depressed nerve function are associated with a breakdown of phosphocreatine.[1] In nerve cells in which chromatolysis has been produced by axon section a markedly increased acid phosphatase activity has been found. This increased activity was proportional to the degree of chromatolysis and was probably associated with increased nucleoprotein synthesis or degradation.[2]

In regenerating neurons which are resistant to the infection with the poliomyelitis virus a decrease of phosphocreatine could also be shown. This change together with the reduction of cytochrome oxidase and succinic dehydrogenase activity in regenerating neurons resembled the effect of cyanide on brain metabolism.[3] Changes in the acid soluble phosphorus compounds in the brain in poliomyelitis have been described, consisting in a greatly increased content of adenosine-triphosphate and a markedly decreased content of phosphocreatine and residual organic phosphate.[4]

Experiments. In the present studies phosphorus-containing compounds were determined in normal mouse brain tissue and compared with those found in the brain of animals infected with polioencephalitis. Five weeks old mice of the C_3H strain received from the colony of Dr. John J. Bittner, University of Minnesota, were inoculated intraperitoneally with the MM poliomyelitis virus. At the height of the infection the animals were decapitated, the brain rapidly removed and homogenized in cold 5% trichloracetic acid. Phosphorus compounds were determined by the method of Schneider[5] which allows for

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