### COMPLEMENT-FIXING ANTIBODIES FOR R. RICKETTSII

# IN SERUMS OF BLACK-TAILED JACK RABBITS

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THE ETIOLOGICAL agent of Rocky Mountain spotted fever, *Rickettsia rickettsii*, is transmitted among mammals by ticks. The organism is present in numerous species of ticks including the rabbit tick *Haemaphysalis leporis-palustris* which is widely distributed throughout the United States (1). Antibodies for *R. rickettsii* may be found in rabbits infested with *Dermacentor andersoni*, *D. variabilis*, *D. parumapertus*, *Amblyomma americanum*, and perhaps other species of ticks (personal communication from Dr. David B. Lackman).

Although the spread of this infection from hares to man is improbable (1-3), the presence of antibodies against *R. rickettsii* in the serums of hares is an indication that the organism is in the area. Infested hares could serve as a reservoir for the infection of additional ticks.

Positive results for antibodies against R. rickettsii have been obtained by other investigators. Philip and associates (4) found complement-fixing antibodies for Rocky Mountain spotted fever in 35 of 260 serums of northern

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This paper is a portion of a thesis submitted by Mr. Pagan. Support for the study was received under Contribution No. 367 of the department of bacteriology, Kansas Agricultural Experiment Station, Manhattan. Additional support was received from the National Institutes of Health, Grant E-1676, and the Kansas Forestry Fish and Game Commission. Nevada jack rabbits. Lechleitner (3) found specific complement-fixing antibodies for R. rickettsii in 10 of 142 serums tested. Stoenner and associates (5), studying the fauna of the Great Salt Lake Desert in Utah, showed that 193 of 773 jack rabbits possessed antibodies against R. rickettsii.

Since the black-tailed jack rabbit, Lepus californicus melanotis, Mearns, is numerous near Lakin, Kans., serums of animals from that area were examined for complement-fixing antibodies against R. rickettsii.

#### **Materials and Methods**

The serum samples were collected monthly from September 1957 through May 1960, as described previously ( $\beta$ ). For this study the method of Bengston (7) was used to test samples for complement-fixing antibodies. Two antigens, supplied by Dr. David B. Lackman, Rocky Mountain Laboratory, Public Health Service, Hamilton, Mont., were used. Spotted fever antigen No. 672 was used in a dilution of 1:4, and spotted fever antigen HLP 664, in a dilution of 1:8.

The antisheep hemolysin and the lyophylized complement used throughout the study were obtained from the Denver Serum Co., Denver, Colo. A 2 percent suspension of sheep red blood corpuscles was prepared each day from blood collected aseptically from the external jugular vein, defibrinated by shaking with glass beads, and stored in a refrigerator at 6-8° C.

For the antigen titration and complement fixation tests, two full units of complement, diluted to be contained in 0.2 ml., were employed. The antigen was titrated against a standard positive serum with a titer of 1:128.

#### Results of tests for Rocky Mountain spotted fever complement-fixing antibodies in 549 serum samples from black-tailed jack rabbits, September 1957 through May 1960

	1	1		
Date collected	Number samples tested	Number positive	Titer of positive	Titer of positive <sup>1</sup>
1957				
Sept Oct Nov	24 13 15	$\begin{array}{c} 1\\ 0\\ 2\end{array}$	1:32 1:8	1:192 1:24
Dec	9	0	1:16	
1958				
Jan Feb Mar Apr	7 15 16 29	0 1 1 4	$1:8 \\ 1:6 \\ 1:12 \\ 1:4 \\ 1:8 \\ 1:4$	$\begin{array}{c} 1:48\\1:24\\1:48\\1:32\\1:32\\1:16\end{array}$
May June July	27 22 26	$\begin{array}{c} 0\\ 1\\ 3\end{array}$	$1:16 \\ 1:8 \\ 1:16$	1:64 1:48
Aug Sept Oct Nov Dec	24 21 28 22 23	1 1 0 0 2	1:8 1:16 1:4 1:32 1:16	$ \begin{array}{c c} 1:32\\ 1:96\\ 1:24\\ \hline \\ 1:96\\ \hline \\ 1:96\\ \end{array} $
1959				
Jan Feb Apr May June July Aug Sept Oct	9 22 22 22	0 0 0 1 0 0 0 0 0 1 2	1:4 	1:24  1:24  1:24 1:24 1:24
Nov Dec			1:8	1:24
1960 Jan Feb Mar Apr May	$\begin{array}{c}2\\15\\0\\10\\16\end{array}$	0 0 0 1 3	1:8 1:16 1:16 1:8	1:48 1:48 1:48
Total	549	26		-
	1	1	1	1

<sup>1</sup> Obtained by Dr. David B. Lackman using the method proposed by Osler and associates (8).

The antigenic unit was considered to be the smallest amount of antigen giving a 4+ fixation with a 1:64 dilution of the positive serum. The usual controls for the complement fixation test were included with each day's tests.

#### Results

During the study period, 549 serum samples were collected from black-tailed jack rabbits. Twenty-two of the 26 serums found to have a titer of 1:4 or greater were sent to Dr. Lackman and were tested by him for antibodies for Rocky Mountain spotted fever. He reported complement-fixing antibodies in these serums as shown in the table. Higher serum titers were obtained by Lackman using the procedure for complement fixation proposed by Osler, Strauss, and Mayer (8). A titer of 1:16 or greater was considered to be significant when this method was employed. The positive serums were also checked for specificity by using typhus antigen. No reaction was obtained against it.

#### Discussion

The 26 serums found to be significantly positive represent 4.8 percent of the total tested. The greatest number of positive serums was found in April, May, July, and September following peak infestations with ticks. Similar results were obtained by Lechleitner ( $\mathcal{I}$ ) who found the greatest number of positive serums during March, April, and September.

The complement-fixing antibodies present in these serums may be assumed to have been formed following infection of the jack rabbits by the feeding of infected rabbit ticks. Although the rabbit tick H. leporis-palustris ordinarily does not parasitize man, the finding of antibodies against R. rickettsii is a significant indicator that R. rickettsii is present in the area.

### Summary

From September 1957 through May 1960, 549 serum samples were collected from black-tailed jack rabbits in the area of Lakin, Kans. The serums were tested for complement-fixing antibodieș against *Rickettsia rickettsii*. Specific antibodies for Rocky Mountain spotted fever were found in 26 of the samples.

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# 1961 Health Trends

Total Federal expenditures for scientific research and development were estimated at \$8.2 billion for fiscal year 1961, an increase of \$400 million over the previous year, according to the 1961 edition of "Health, Education, and Welfare Trends."

Included in the data presented are:

• Of 93,000 deaths from accidents in 1960, some 37,000 were attributed to motor vehicles. For every 10,000 persons, more than 2 die from motor vehicle accidents in any given year.

In 1960 the index of all medical care prices averaged 156.2 (1947-49=100). Medical care prices have increased rapidly since World War II with the largest increases occurring in hospital room rates, group hospitalization premiums, and physicians' and dentists' fees.
In 1959 the private expenditures for medical care and voluntary health insurance totaled

\$18.3 billion, or 5.4 percent of disposable personal income. Of the per capita expenditure of \$105, \$32 was spent for hospital services, \$29 for physicians, \$27 for medicines and appliances, \$11 for dentists' services, and \$6 for all other medical expenses.

• Since 1931 the use of general and special hospitals has increased markedly, total days in the hospital having risen 46 percent. Although the rate of admissions to these hospitals has increased, the average length of stay has declined.

• At the end of 1960, 25 years after the initiation of the social security system, there were 14.8 million beneficiaries under the Old-Age, Survivors, and Disability Insurance Program.

Copies of "Health, Education, and Welfare Trends" may be purchased at \$1 per copy from the Superintendent of Documents, U.S. Government Printing Office, Washington 25, D.C.