INCIDENCE OF REACTIVE VDRL TESTS IN THE NORMAL RABBIT*†

BY

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Experimental ocular and neurosyphilis has been investigated in this laboratory during the past 3 years in three species—rabbits, owl monkeys, and squirrel monkeys. Rabbits were selected as a control to the primate experiments because of the large available literature concerning experimental syphilis in the rabbit. During the course of these studies, it became apparent that many rabbits purchased commercially and in apparent good health were found to have reactive serum reagin tests before their use for any experiments whatever. Review of the literature revealed that such a phenomenon had been reported by Kolmer and Casselman (1913). This paper presents the findings of a study made in order to determine the incidence of reactive VDRL tests in the normal rabbit population.

Material and Methods

Three groups of rabbits were studied.

1) 42 rabbits used in the past 3 years All of these animals were purchased commercially, were in apparently good health, and were housed in individual cages. Blood specimens were drawn before experimental use of the animals and submitted to the Venereal Disease Research Laboratory in Atlanta, Georgia, where all the serological tests were performed. Data on this group were as follows:

<p>| TABLE I |
| RESULTS OF PRE-INOCULATION SERUM VDRL TEST |</p>
<table>
<thead>
<tr>
<th>No. of Rabbits</th>
<th>Reactive</th>
<th>Weakly Reactive</th>
<th>Non-reactive</th>
<th>Per cent. Reactive</th>
</tr>
</thead>
<tbody>
<tr>
<td>42</td>
<td>5</td>
<td>6</td>
<td>31</td>
<td>11/42 = 26 per cent.</td>
</tr>
</tbody>
</table>

2) 24 rabbits in apparent good health All were purchased at various times during the past year from the same local breeder. However, in this group, all VDRL tests were performed in this laboratory. The results were as follows:

<p>| TABLE II |
| RESULTS OF PRE-INOCULATION SERUM VDRL TEST |</p>
<table>
<thead>
<tr>
<th>No. of Rabbits</th>
<th>Reactive</th>
<th>Weakly Reactive</th>
<th>Non-reactive</th>
<th>Per cent. Reactive</th>
</tr>
</thead>
<tbody>
<tr>
<td>24</td>
<td>0</td>
<td>6</td>
<td>18</td>
<td>6/24 = 25 per cent.</td>
</tr>
</tbody>
</table>

By combining the weakly reactive and reactive tests in groups 1 and 2, it was evident that one-fourth of apparently healthy rabbits showed reactive VDRL tests. Also, there was excellent correlation between the findings in two different laboratories.

Because of this observation, a field trip to the local rabbitry was made by two of us (J.S.P. and M.A.R.). This institution housed approximately 500 rabbits of various ages and in apparent good health.

3) A random selection of 83 animals made on the basis of age Blood tests were drawn using sterile precautions from the car veins. All the serum VDRL tests were performed in this laboratory. The results are shown in Table III (opposite).

Results

Considering the data of all three groups of animals, of 149 rabbits tested, strongly reactive VDRL tests were found in fifteen (10 per cent.), and weakly reactive in 45 others (30 per cent.). Combining the reactive and weakly reactive tests, a positive reagin test was found in sixty (40 per cent.) of all rabbits surveyed.

Discussion

The finding that more than one out of four commercially purchased rabbits in apparent good health showed a reactive serum VDRL test was
VDRL TESTS IN THE NORMAL RABBIT

TABLE III
RESULTS OF SERUM VDRL TEST

<table>
<thead>
<tr>
<th>No. of Rabbits</th>
<th>Approximate Age</th>
<th>Reactive</th>
<th>Weakly Reactive</th>
<th>Non-reactive</th>
<th>Per cent. Reactive</th>
</tr>
</thead>
<tbody>
<tr>
<td>12</td>
<td>8-12 wks</td>
<td>1</td>
<td>4</td>
<td>7</td>
<td>5/12 = 42 per cent.</td>
</tr>
<tr>
<td>6</td>
<td>6-12 mths</td>
<td>3</td>
<td>1</td>
<td>2</td>
<td>4/6 = 67 per cent.</td>
</tr>
<tr>
<td>65</td>
<td>over 12 mths</td>
<td>6</td>
<td>28</td>
<td>31</td>
<td>34/65 = 52 per cent.</td>
</tr>
</tbody>
</table>

There are various possible explanations for such a finding. The fact that rabbits harbour an endemic spirochaetal disease—venereal spirochaetosis—due to *Treponema cuniculi*, an organism morphologically indistinguishable from *T. pallidum*, must be considered. Arzt and Kerl (1914), who studied a total of 853 rabbits in an attempt to determine the incidence of venereal spirochaetosis, noted flat, scaly anogenital lesions containing numerous spirochaetes in 26-9 per cent. of their adult rabbits. Noguchi (1921, 1922) reported the incidence of infection in American rabbits to be between 10 and 30 per cent. Adams, Cappell, and McCluskie (1928) stated that, according to reputable observers, 20 to 40 per cent. of the wild rabbits in England were infected with *T. cuniculi*. Fried and Orlov (1932) reported on an epidemic of rabbit spirochaetosis in Moscow, and found that 704 of 3,650 rabbits (19·3 per cent.) had the spontaneous disease. They considered age to be an important factor in rabbit spirochaetosis. Thus, of 2,893 animals aged one year or above which had mated, 1,304 (46·4 per cent.) were infected, but of 5,800 rabbits less than 8 months old most of which had not yet mated, only 40 (0·7 per cent.) were said to be diseased.

The world-wide distribution of rabbit spirochaetosis and the recurrent nature of figures of incidence similar to the findings in this study are interesting. Such figures have not been universal, however. McLeod and Turner (1946, a, b) stated that only six out of 1,800 rabbits used in their laboratory over a 3-year period were infected with venereal spirochaetosis. An important consideration in this regard is whether or not rabbits can harbour this infection in a clinically inapparent or latent state. The occurrence of clinically inapparent spirochaetosis in the rabbit was first experimentally demonstrated by Bessemans and De Wilde (1937).

Other factors should be considered in evaluating the role of *T. cuniculi* as a possible cause for the serological phenomenon here reported. Thus, TPI and FTA-ABS tests were performed on thirty of the rabbits whose sera were studied in Atlanta. Of this number, the TPI and FTA-ABS were non-reactive in 28, but two rabbits were of interest.

Rabbit 93 had a weakly reactive FTA-ABS test on pre-inoculation blood drawn on January 1, 1964. The TPI at that time was non-reactive. This rabbit was then inoculated with *T. pallidum*, and subsequently had several reactive FTA-ABS tests, which would not be unexpected after syphilitic infection.

Rabbit 649 showed a weakly reactive FTA-ABS test on July 6, 1964, but repeated FTA-ABS tests on August 6, 1964, June 2, 1965, and August 2, 1965, were non-reactive.

Thus, TPI and FTA-ABS tests were non-reactive in 28 of thirty rabbits before inoculation, although ten of these animals showed reactive serum VDRL tests.

Two other points merit consideration:

1. The same local breeder has for several years provided all rabbits used at the University of Miami. At varying intervals during a 3-year period, 26 per cent. of 42 rabbits were found to have reactive VDRL tests. A similar finding was noted in 25 per cent. of another 24 rabbit sera tested in our laboratory during the past year. Because of the relative length of time involved, and the excellent correlation between two laboratories, the finding of a value approximately twice this level at the time of the field trip would raise the question of an undetected spirochaetosis epidemic in the rabbitry.

2. The great prevalence of weakly reactive VDRL tests. Of the sixty reactive tests, 45 were weakly reactive. The importance of dilutional factors was emphasized by Kolmer and Casselman (1913). The cause of the serological phenomenon here reported could be due to biochemical mechanisms other than previous or subclinical *T. cuniculi*.
infection. The non-reactive TPI and FTA-ABS tests found in 28 rabbits, ten of which had a reactive VDRL, should be noted. Certainly, the occurrence of reactive reagin tests in the rabbit is as yet unexplained, and it is hoped that this report will stimulate the serological investigation of other large series of rabbits of various ages to help explain the pathogenesis of this phenomenon. The significance of the observation for experimental syphilis research in the rabbit is obvious.

Summary
The serum VDRL test, studied in 149 apparently healthy rabbits, was reactive in 10 per cent., and weakly reactive in 30 per cent. The importance of further studies to document this finding and to determine its pathogenesis and significance for syphilis research in the rabbit is evident.

REFERENCES

L’incidence des tests positifs VDRL chez le lapin normal
Résumé
Le sérum de 149 lapins apparentemment sains soumis au test VDRL a montré une réaction positive chez 10 pour cent de ces animaux et une réaction positive faible chez 30 pour cent. L’importance d’études additionnelles pour documenter cette conclusion et pour déterminer sa pathogénèse et sa signification dans les travaux de recherches au sujet de la syphilis chez le lapin est évidente.
Incidence of reactive VDRL tests in the normal rabbit.

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