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QUANTITATIVE ASPECTS OF LEPROSY IN ARMADILLOS W. F. Kirchheimer and R. M. Sanchez U.S. Public Health Service Hospital, Carville

In 1971 Kirchheimer and Storrs (1) first reported lepromatoid leprosy in an experimentally infected armadillo. Skin lesions during life yielded ten billion (10^{10}) <u>M</u>. <u>leprae</u> per gm.

In their detailed histopathologic and bacteriologic autopsy report of this armadillo, Kirchheimer et al. (2) showed that disseminated leprosy in the armadillo in contrast to man involves all organs, even the lung and meninges. They also showed that the <u>M</u>. <u>leprae</u> in organs and lepromas are viable and far more numerous than in man. Numbers of leprosy bacilli recovered from 1 gm of the different tissues of two different armadillos are shown in Tables 1 and 2.

Table 1. Number of <u>M</u>. <u>leprae</u> per gram of autopsy material of the first leprous armadillo

Right earlobe	$2.0 \times 10^{10}(7)$ *
Lymph node (groin	$8.9 \times 10^9(15)$ $6.6 \times 10^2(21)$
Spleen	$6.6 \times 10^{9}(21)$
Brain	$6.2 \times 10^{5}(38)$
Uninoculated skin	$6.2 \times 10^{5}(38)$ 1.1 × 10 ⁹ (12)
Lung	$3.9 \times 10^8(12)$ 2.2 × 10 ⁸ (33)
Liver	$2.2 \times 10^{\circ}(33)$

%Numbers in parentheses are Morphological Indices. Kirchheimer, Storrs and Binford, Int. J. Lep. <u>40(3)</u>:229-242, 1972. Table 2. Numbers of M. leprae per gm of tissue of armadillo 34

Tissue	Number of <u>M</u> . <u>leprae</u>			
Skin at site of inoculation Skin (uninoculated) Lymph node (inguinal) Leproma (left thigh) Liver Spleen	$3.6\pm0.7 \times 10^{8}(15) \times 9.5\pm0.1 \times 10^{7}(16) \times 10^{7}(16) \times 10^{10}(18) \times 10^{10}(18) \times 10^{10}(21) \times 10^{9}(21) \times 10^{9}(21) \times 10^{9}(21) \times 10^{8}(12)$			

*Numbers in parentheses are percentages of solidly staining rods (Morphological Index). Yoshizumi, Kirchheimer and Asbury, Int. J. Lep. <u>42</u>(3):252-259, 1974.

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Table 3 shows the means and their standard deviation of the bacterial numbers recovered from 1 gm of different livers, spleens, lymph nodes and cutaneous lepromas. As one would expect for various reasons there are great variations in the number of \underline{M} . <u>leprae</u> recovered.

Table 3. Means of <u>M</u>. <u>leprae</u> counts per gm of tissue from different armadillos at Carville

Tissue	Mean Count	Standard Deviation
Liver	2.25×10^9 1.90×10^9 1.80×10^{10} 1.90×10^{10}	$\pm 2.10 \times 10^{9}$
Spleen	1.90×10^{9}	$\frac{1}{2} \cdot 10 \times 10^{9}$ $\frac{1}{2} \cdot 10 \times 10^{10}$ $\frac{1}{2} \cdot 10^{10} \times 10^{10}$
Lymph node	1.80×1010	$\pm 1.90 \times 10^{10}$
Leproma	1.90×10^{10}	$\pm 6.25 \times 10^{10}$

Percentages of armadillos developing disseminated leprosy depend on the infectious dose. Kirchheimer and Sanchez (3) have stated that statements with precise meaning require experiments with different dilutions from the same suspension inoculated by the same route at the same time. The results of an experiment where 15 adult armadillos were infected intracutaneously with 10^7 <u>M</u>. <u>leprae</u> from the same suspension at the same time are shown in Table 4.

Table 4. Leprosy in armadillos 1095 days after intracutaneous infection with 107 <u>M</u>. <u>leprae</u> of human origin

		<u>M</u> . <u>leprae</u> seen at infection site		Viable <u>M</u> . <u>leprae</u> in infection site			
		Positive	Negative	Positive	Negative	Positive	Negative
Deaths	5	4	1	4	1	5	0
Survivors	10	4	6	4	6	4	6
Totals	15	15		15		15	

Five of the 15 armadillos were dead from disseminated leprosy within 1095 days. (Range of death-times 397 days to 1016 days). Four of the survivors had some sign of dissemination. Six had none. We do not know whether the infection can come to a standstill in some

With increasing challenge mean death times decrease and percentages of death increase. With intracutaneous injection of from 107 to 7.8 x 107 acid-fast particles, the mean death time was 802 days. With 2.0 to 7.3 x 108 particles it was 617 days with 80 percent deaths after 848 days. The mean amounts of <u>M. leprae</u> at time of death per gm of liver were 5.3×10^9 and 2.6×10^9 respectively (difference not significant).

Neither rate of death, time of death or bacterial numbers in the tissues show that immature armadillos are more susceptible to intradermal challenge than mature ones. This conclusion is based on the following findings. Fourteen armadillos were infected simultaneously by intradermal inoculation of 3.5×10^7 <u>M</u>. <u>leprae</u> of human origin. In the mouse foot-pad 10⁴ <u>M</u>. <u>leprae</u> from this suspension increased to 10^5 in six months. At the time of infection four of these armadillos were 2 months and 12 days old, four were 2 months and 26 days old and three were 4 months old. These eleven armadillos were born in captivity and stem from litters of females fertilized before capture. The remaining three armadillos were fully grown mature animals.

Two of the mature armadillos were sacrificed after 953 and 975 days respectively, because they had signs of disseminated leprosy. One of these armadillos had 10^{10} <u>M</u>. <u>leprae</u> per gm of liver. The other armadillo had 1.3×10^9 <u>M</u>. <u>leprae</u> per gm of liver and 8.2×10^8 per gm of spleen. The third adult armadillo had died with disseminated leprosy 792 days after infection. This animal had 2.6 $\times 10^{10}$ <u>M</u>. <u>leprae</u> per gm of spleen.

Three of the immature armadillos had to be sacrificed 4 to 5 months after experimental infection because of injuries. No <u>M</u>. <u>leprae</u> were found on microscopic examination in any of their organs. The armadillo which was sacrificed after 4 months had 2.0×10^5 <u>M</u>. <u>leprae</u> per gm of tissue in the site of infection. One of the remaining eight young armadillos died with disseminated leprosy after 768 days. An additional three young armadillos were sacrificed 762, 954 and 1040 days after infection because they had disseminated leprosy. The armadillo sacrificed after 954 days had only 3.9 x 10⁷ <u>M</u>. <u>leprae</u> per gm of liver and 1.1 x 10⁸ per gm of spleen.

Four young armadillos are still alive 1050 days after infection. Three of these have no signs of dissemination. The fourth young armadillo has <u>M</u>. <u>leprae</u> in the skin of the outer ear and in the blood.

REFERENCES

- Kirchheimer, W. F. and Storrs, E. E. Attempts to establish 1. the armadillo (Dasypus novemcinctus Linn.) as a model for the study of leprosy. I. Report of lepromatoid leprosy in an experimentally infected armadillo. Int. J. Lep. 39(3):693-702, 1971.
- Kirchheimer, W. F., Storrs, E. E. and Binford, C. H. Attempts 2. to establish the armadillo (<u>Dasypus</u> novemcinctus Linn.) as a model for the study of leprosy. II. Histopathologic and bacteriologic post-mortem findings in lepromatoid leprosy in an armadillo. Int. J. Lep. 40(3):229-242, 1972.
- 3. Kirchheimer, W. F. and Sanchez, R. M. Survival of Mycobacterium leprae in cutaneous inoculation sites of armadillos. Lep. in India 47(1:5-8, 1975.