Mycobacterium lepraemurium Infection of Nude Athymic (nu/nu) Mice

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Nude athymic (nu/nu) mice on a BALB/c background and their heterozygous euthymic litter mates (nu/+), were infected with either 10^6 or 10^4 Mycobacterium lepraemurium organisms intravenously or in the left hind footpad (LHF). After LHF infection with 10^6 M. lepraemurium organisms, nu/+ mice slowly developed a response that consisted of LHF swelling and local resistance to Listeria monocytogenes. The lower inoculum induced a proportionately lower response in nu/+ mice, but the nu/nu mice developed neither LHF swelling nor resistance to L. monocytogenes in response to either dose of M. lepraemurium. Counts of M. lepraemurium in the LHF revealed no difference between the nu/+ and nu/nu mice. After intravenous infection the nu/+ mice developed splenomegaly, but did not otherwise differ from nu/nu mice with respect to resistance to intravenous challenge with L. monocytogenes or growth of M. lepraemurium in the spleen. In light of the poor responsiveness of nu/+ mice in this experiment, they were then compared with CB6 and B6D2 mice, which are genetically susceptible and resistant to M. lepraemurium, respectively. These mice were infected with either 10^6 or 10^4 M. lepraemurium cells or 10^6 Mycobacterium bovis BCG cells in the LHF. Once again the nu/+ mice responded poorly to M. lepraemurium, the B6D2 mice gave an intermediate response with respect to LHF swelling and resistance to L. monocytogenes. However, M. lepraemurium grew to higher numbers in the LHF of nu/+ and CB6 mice than in B6D2 mice, revealing, in CB6 mice, a dissociation between resistance to L. monocytogenes and M. lepraemurium. All three mouse strains responded strongly to M. bovis BCG, but there was a suggestion that nu/+ mice might be more susceptible to this agent than the other two strains. I concluded that the failure of nu/+ mice to restrict the growth of M. lepraemurium more than nu/nu mice was due to the intrinsic genetic susceptibility of both types of mice. In effect, the nu/+ mice behaved like nu/nu mice, as if they too were deficient in T lymphocytes that were responsive to M. lepraemurium.

Mycobacterium lepraemurium infection of mice has been studied by several groups of investigators as a model of human lepromatous leprosy (2, 4, 13). Genetically resistant and susceptible strains of mice have been identified (4, 8, 13), yet even in the resistant strains of mice intravenous (i.v.) infection is invariably fatal. However, some investigators have found that resistant mice can survive footpad infection with moderate numbers of M. lepraemurium cells (3). Resistant strains and some, but not all, susceptible strains of mice develop cell-mediated immunity (CMI) to M. lepraemurium, particularly after footpad inoculation (5, 13, 19). Such CMI is expressed as delayed hypersensitivity (19), nonspecific resistance to a heterologous organism such as Listeria monocytogenes (13), and cross-reactive resistance to other mycobacteria species, e.g., Mycobacterium bovis BCG and Mycobacterium tuberculosis R1Rv (12). It has even been found that adaptive immunity to the homologous organism can be expressed in sublethally irradiated recipient mice (10). Despite these data, it has not been resolved whether the CMI response that is generated in infected mice has any measurable effect in restraining the in vivo replication of M. lepraemurium.

An earlier study from my laboratory revealed that in the initial stages of i.v. infection of genetically resistant mice, the B6D2F1 hybrids, the replication of M. lepraemurium bacilli was restrained during a period when strong delayed hypersensitivity was elicitable. Later, there was a decay of hypersensitivity and concomitant accelerated multiplication of M. lepraemurium bacilli (19). The weakness of those data is that it is uncertain whether the initially poor replication of M. lepraemurium was caused by the induction and expression of CMI or was a result of the lag period that commonly follows the inoculation of bacteria into an animal, or even into fresh culture medium in vitro.

One approach to the resolution of this problem is to compare the growth of the pathogen in normal mice and in those that are depleted of T lymphocytes. If CMI were to effectively restrain the multiplication of the pathogen in the former mice, then higher numbers of microorganisms would be recovered from the latter. The results of such experiments with homozygous athymic nude mice and their corresponding heterozygous littermates, which possess thymus glands and are immunologically normal, are now presented.

MATERIALS AND METHODS

Animals. Homozygous, nude, athymic (nu/nu) mice and heterozygous, euthymic (nu+/+) littermates were obtained from Charles River Breeding Laboratories, Wilmington, Mass. These were albino mice with the nu mutation on a BALB/c background. CB6F1 (BALB/cJ × C57BL/6J) and B6D2F1 (C57BL/6J × DBA/2J), which are genetically susceptible and resistant to M. lepraemurium, respectively, were purchased from Jackson Laboratories, Bar Harbor, Maine. Only female mice were used; they were introduced into experiments at approximately 2 months of age. In the text these mice will be designated as nu/nu, nu/+, CB6, and B6D2, respectively.

Microorganisms. M. lepraemurium bacilli were passaged in CB6 mice; organisms were recovered from the liver and spleen, purified, and stored at −70°C (13). M. bovis BCG Pasteur (TMC 1011) was grown in a modified Proskauer and Beck medium and stored at −70°C. A mouse-virulent strain...
of *L. monocytogenes* was grown in tryptic soy broth and also stored at -70°C. These stored cultures facilitated the use of the same bacterial stocks and relatively uniform inocula throughout the entire series of experiments.

**Infection of mice.** Mice were infected by injection into either the lateral tail vein (i.v.), in a 0.2-ml volume, or the left hind footpad (LHF), in a 0.04-ml volume. The presumptive approximate inocula were as follows: 10⁶ or 10⁸ *M. lepraemurium* bacilli, given i.v. and into the LHF; 10⁸ *M. bovis* BCG bacilli, given into the LHF; and 10⁵ *L. monocytogenes* bacilli, given i.v. and into the LHF.

**Counts of bacteria.** The appropriate organs, spleens or LHF, were removed and homogenized in sterile saline with Potter-Elvehjem and VirTis homogenizers, respectively. For total counts of *M. lepraemurium* bacilli, Reich slide smears were prepared (10, 22), stained with phenol auramine, and counted with a fluorescence microscope. Viable counts of *M. bovis* BCG and *L. monocytogenes* bacilli were made by inoculating Middlebrook 7H-10 agar or tryptic soy agar plates, respectively, with 0.1-ml volumes of suitably diluted homogenates. After appropriate periods of incubation at 37°C, the colonies were counted.

**Nonspecific resistance.** Mice were challenged either i.v. or into the LHF with 10⁵ *L. monocytogenes* organisms. After 24 h, the mice were killed; either the spleen or LHF was removed, as appropriate, and homogenized, and viable counts were made.

**Spleen weight.** Spleens that were removed for counts of *M. lepraemurium* bacilli were first weighed.

**Footpad swelling.** The thickness of each hind foot was measured with dial-gauge calipers to the nearest 0.1-mm unit. Swelling of the LHF was obtained by subtracting the right hind foot measurement from that of the LHF.

**Data analysis.** In all experiments there were five mice per group, but there were occasional missing data because of unanticipated death of mice or technical problems. The viable counts were transformed to log₁₀, and these data were evaluated by two-way analysis of variance. Group geometric means were compared by using the Scheffé test (23). This is a stringent test for which a value of *P* < 0.05 is considered sufficient. Consequently, none of the differences were tested for lower levels of probability. The *M. lepraemurium* and *M. bovis* BCG data were expressed as geometric absolute counts. Resistance to *L. monocytogenes* was expressed by subtracting the geometric mean viable count of the control group from the corresponding counts from test groups in the same experiment (Table 1).

The footpad swelling measurements were converted to square roots, since this transformation permitted the legitimate use of parametric statistical methods. Statistical analyses were performed as described above, but the mean values were reconverted to standard arithmetic units for data presentation in the charts. The spleen weight data were analyzed in standard arithmetic units.

**RESULTS**

Response of *nu/+* and *nu/nu* mice to footpad infection with *M. lepraemurium*. Mice were infected with either a high dose (10⁶ organisms) or a moderate dose (10⁵ organisms) of *M. lepraemurium* bacilli in the LHF. Footpad swelling was measured weekly for 8 weeks and then every 2 weeks until the experiment was terminated after 24 weeks. No swelling was observed in any group of mice until week 8, when the site of inoculation of 10⁶ *M. lepraemurium* cells into *nu/+* mice became swollen. Thereafter, those feet continued to swell as shown in Fig. 1 (for simplicity, only measurements made every 4 weeks are illustrated). The LHF of *nu/+* mice inoculated with 10⁶ mycobacteria first became swollen at week 12. The LHF of *nu/nu* mice infected with 10⁵ bacilli never became swollen, and the feet inoculated with 10⁸ bacilli were swollen only at 16 weeks. No nude mice survived beyond 16 weeks, but death should not be attributed to *M. lepraemurium*, since uninoculated control *nu/nu* mice also began to die 12 to 16 weeks into the experiment.

Resistance to *L. monocytogenes* in the LHF was estimated at 4-week intervals (Fig. 2). By week 4, even before measurable swelling of the foot had occurred, *nu/+* mice infected with 10⁸ *M. lepraemurium* organisms were highly resistant to *L. monocytogenes*. Thereafter, resistance steadily declined, but was still statistically significant (*P* < 0.05) through week 20. *nu/+* mice infected with 10⁶ *M. lepraemurium* organisms were significantly resistant (*P* < 0.05) only at week 16. Increased local resistance to *L. monocytogenes* was not expressed by *nu/nu* mice that had been infected with either dose of *M. lepraemurium*. Indeed, at week 12 these mice permitted enhanced growth of *L. monocytogenes* in the LHF (*P* < 0.05).

Counts of acid-fast bacilli in the LHF were made at 4-week intervals (Fig. 3). It is apparent that there was no material difference in the replication rates of *M. lepraemurium* in the LHF of *nu/+* and *nu/nu* mice, despite the

### TABLE 1. Resistance to *L. monocytogenes* in spleen after i.v. infection with *M. lepraemurium*

<table>
<thead>
<tr>
<th>Week</th>
<th>M. lepraemurium infection dose (cells)</th>
<th>Viable <em>L. monocytogenes</em> cells/spleen (log₁₀)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td><em>nu/+</em> mice</td>
</tr>
<tr>
<td>4</td>
<td>0</td>
<td>6.16 ± 0.32</td>
</tr>
<tr>
<td></td>
<td>10⁶</td>
<td>6.17 ± 0.32</td>
</tr>
<tr>
<td></td>
<td>10⁸</td>
<td>5.98 ± 0.52</td>
</tr>
<tr>
<td>8</td>
<td>0</td>
<td>5.55 ± 0.48</td>
</tr>
<tr>
<td></td>
<td>10⁶</td>
<td>5.48 ± 0.48</td>
</tr>
<tr>
<td></td>
<td>10⁸</td>
<td>5.13 ± 0.46</td>
</tr>
<tr>
<td>12</td>
<td>0</td>
<td>5.19 ± 0.28</td>
</tr>
<tr>
<td></td>
<td>10⁶</td>
<td>4.71 ± 0.66</td>
</tr>
<tr>
<td></td>
<td>10⁸</td>
<td>4.77 ± 0.65</td>
</tr>
</tbody>
</table>
marked differences in footpad swelling and resistance to *L. monocytogenes*. The growth rate of *M. lepraemurium* was inversely related to the inoculum size in both nul+ and nu/nu mice, a phenomenon that has also been observed in BCG (9) and *M. leprae* (21) infection and has previously been ascribed to the more rapid development of CMI in animals immunized with large inocula. This explanation cannot validly apply to the nu/nu mouse.

**Response of nul+ and nu/nu mice to i.v. infection with *M. lepraemurium***. Mice were immunized with approximately 10^8 or 10^9 *M. lepraemurium* organisms, and infection was monitored with respect to spleen weight, resistance to *L. monocytogenes* in the spleen after i.v. challenge, and total acid-fast bacilli per spleen. These measurements were made every 4 weeks, until all of the mice of both strains had died by week 16. The spleen weights of control nul+ and nu/nu mice remained fairly steady at approximately 0.2 g throughout the experiment (Fig. 3). Between 4 and 12 weeks there was steady enlargement of spleens of nul+ mice infected with 10^8 *M. lepraemurium* organisms, reaching a maximum value of 0.74 g at 12 weeks. Those nu/nu mice that were infected with the larger dose of mycobacteria also exhibited splenic enlargement, 0.37 g, at 8 weeks, but not at 12 weeks. The result at 12 weeks is based on only three surviving mice that were moribund. The nul+ mice infected with 10^6 bacteria demonstrated splenomegaly only at week 12, and no splenomegaly was observed in nu/nu mice infected with that inoculum.

Neither the *M. lepraemurium*-infected nul+ mice nor the nu/nu mice were significantly resistant to *L. monocytogenes* at any time point. These results are in part attributable to the unusually large intragroup variation in counts, as signified by high standard deviations (Table 1). The resistance of even control mice to *L. monocytogenes* was unusually variable, and the resistance of control nu/nu mice was in general higher than that of nul+ mice (7, 16), reaching a level of statistical significance (*P* < 0.05) at week 4, but not at weeks 8 and 12. Finally, the growth of *M. lepraemurium* was closely similar in the spleens of nul+ and nu/nu mice (Fig. 3). Once again the bacilli multiplied more freely from the smaller inocula, and by week 12 the original disparity between the inocula, approximately 100-fold, had almost disappeared.

**Response of B6D2, CB6, and nul+ mice to *M. lepraemurium* and *M. bovis* BCG**. The previous experiments conveyed the impression that responsiveness to *M. lepraemurium* in the immunologically competent nul+ mice was low, even after footpad inoculation, a route that favors the induction of strong CMI. An experiment was therefore designed to compare the response of nul+ mice with those of CB6 and B6D2 mice, which are susceptible and resistant to *M. lepraemurium*, respectively (13), and to compare the responsiveness of these mouse strains to *M. lepraemurium* and another mycobacterial species, *M. bovis* BCG.

Mice of each of the three strains were divided into four groups, one of which was set aside for uninfected controls. The other three groups were infected in the LHF with approximate inocula of either 10^8 live *M. lepraemurium* ...
organisms, 10^6 live *M. lepraemurium* organisms, or 10^6 live *M. bovis* BCG organisms. At weekly intervals for 8 weeks, the infected and control mice were tested for swelling of the LHF and resistance to *L. monocytogenes* at that site. In addition, total counts of acid-fast bacilli were made from the LHF of mice infected with *M. lepraemurium* at weeks 4 and 8; weekly viable counts of BCG were made from the LHF of mice infected with that organism.

The footpad swelling and *L. monocytogenes* resistance data are illustrated in Fig. 6, 7, and 8. The response of mice to the higher dose of *M. lepraemurium* is shown in Fig. 6. There was a latent period of 2 weeks during which there was neither swelling of the LHF nor increased resistance to *L. monocytogenes*. These and other negative data are omitted from Fig. 6, 7, and 8. There was appreciable LHF swelling of similar magnitude in all mouse strains at week 3. Thereafter, swelling of the LHF in CB6 and B6D2 mice increased gradually to a level of approximately 1 mm at 8 weeks. By contrast, in *nu/+* mice the LHF swelling did not increase appreciably between weeks 3 and 6, but then increased sharply, exceeding the LHF swelling observed in the other two strains of mice. This pattern of delayed footpad swelling followed by excessive swelling is reminiscent of that observed in mice that are depleted of T lymphocytes (13). Considering now the development of resistance to *L. monocytogenes*, very high levels of resistance were observed in CB6 and B6D2 mice at week 3 and were sustained for the duration of the experiment. Significant resistance (*P* < 0.05) was first observed in *nu/+* mice at 6 weeks. There was a transitory decrease at week 7, probably due to experimental variation, followed by recovery of resistance at week 8, at which time resistance to *L. monocytogenes* was similar in all three strains of mice.

The response to the lower dose of *M. lepraemurium* (Fig. 7) reveals clear differences from the data in Fig. 6. As expected from the use of a smaller inoculum, the latent period between infection and the onset of LHF swelling and resistance to *L. monocytogenes* was prolonged. Significant LHF swelling was seen only in CB6 mice, and then not until week 7 of the experiment. However, these mice developed high resistance to *L. monocytogenes* in the LHF at week 4, which anticipated the footpad swelling by 3 weeks and was sustained. The B6D2 mice showed no significant LHF swelling, yet developed significant (*P* < 0.05) resistance to *L. monocytogenes* by week 5; resistance increased to a level similar to that observed in CB6 mice by week 6 and then remained fairly steady. The *nu/+* mice also failed to develop LHF swelling and did not develop resistance to *L. monocytogenes* until the final time point at week 8.

It might be inferred that *nu/+* mice do not respond strongly to mycobacterial antigens, but this idea is refuted by their response to *M. bovis* BCG (Fig. 8). It is evident that footpad swelling after inoculation of 10^6 *M. bovis* BCG organisms is very small in all three mouse strains (note that the ordinate is in 0.1-mm units), but the response of *nu/+* mice is at least as good as those of the other strains. The
FIG. 8. Swelling and resistance to *L. monocytogenes* (LM) in the LHF after injection of 10^8 *M. bovis* BCG organisms into that site. Symbols are as in Fig. 6. Note that the scale of the ordinate of footpad swelling is 10 times greater than that in Fig. 6 and 7.

The swelling and resistance data are indirect measures of the mycobacterial infection, but the counts of *M. lepraemurium* and *M. bovis* BCG are more pertinent. The counts of *M. lepraemurium* are shown in Fig. 9, which shows that the actual inocula were 3 x 10^7 and 3 x 10^8 bacilli, respectively, substantially lower than the presumptive inocula. There was replication in the LHF of both inocula in all strains of mice between 0 and 4 weeks. At that time the larger inoculum yielded closely similar counts in all of the mouse strains, but the lower inoculum counts were higher in the CB6 mice than in the B6D2 mice (*P < 0.05), with the nul^+^ counts occupying an intermediate position. By 8 weeks, the counts revealed sharper differences between the mouse strains. The larger inoculum yielded higher counts in the nul^+^ mice than in the CB6 mice (*P < 0.05) and higher counts in the CB6 mice than in the B6D2 mice (*P < 0.05).

The smaller inoculum yielded similar counts from the nul^+^ and CB6 mice, which were very much higher, approximately 10-fold, than those obtained from the B6D2 mice (*P < 0.05). The viable counts of *M. bovis* BCG in the LHF are shown in Fig. 10. After inoculation of 10^6 viable *M. bovis* BCG organisms into the footpad there was little or no increase in bacterial members, followed by a long period of declining viable bacilli. With the exception of the week 2 counts, the fate of *M. bovis* BCG bacilli in the LHF of CB6 and B6D2 mice was indistinguishably similar. However, for most of the period of the study higher numbers of viable bacilli were recovered from the LHF of nul^+^ mice than from the other strains. The differences were of the order of 0.3 to 0.5 log_{10} units and were statistically significant (*P < 0.05) at weeks 3 through 7. The upturn of viable bacilli at week 8 in the CB6 and B6D2 mice is attributed to random experimental variation.

DISCUSSION

Before an attempt to interpret the data from these experiments, two methodological problems need to be addressed. The first concerns the use of nulu mice as T lymphocyte deficient animals. The original presumption that these animals completely lacked T lymphocytes has been modified (20), but there is no question that nulu mice are severely depleted of T cells. There is internal evidence of such depletion in this study in that the lack of footpad swelling in response to *M. lepraemurium* infection (Fig. 1) is similar to that previously observed in mice that had been depleted of T lymphocytes by adult thymectomy, lethal irradiation, and bone marrow reconstitution (13). The second problem concerns the counts of *M. lepraemurium* bacilli. We have now accumulated 10 years of experience in preparing and counting stained tissue homogenates. During that time there have been recurrent episodes in which either inexplicably low or high counts occurred. The intrinsic inaccuracy of visual counting of bacteria (15) is compounded by such variables as imperfect homogenization of tissues and the unpredictable acid fastness of mycobacteria (17), leading to apparent disappearance of organisms (14). Consequently, apparent differences between counts at individual time points, even if statistically significant, carry less scientific weight than the
general trend of counts, consolidated from several time points.

With these provisos, it is clear that the response of nulnu mice to *M. lepraemurium* differed from that of their nul+ controls. After footpad infection, the latter developed higher levels of early footpad swelling, indicative of hypersensitivity granuloma formation (5) and increased nonspecific resistance to *L. monocytogenes*. Both swelling and resistance were proportional to the inoculum size, but resistance preceded the swelling. After i.v. infection, splenomegaly was more marked in the nul+ mice, but there was no concomitant increase of resistance of *L. monocytogenes*, so one cannot infer that the splenomegaly was a consequence of the induction of CMI. The evaluation of the i.v. resistance data is complicated by the enhanced systemic resistance of nulnu mice to *L. monocytogenes* (7, 16). However, despite these differences between the nul+ and nulnu mice, closely similar numbers of *M. lepraemurium* bacilli were recovered from them, suggesting that if indeed the nul+ mice did develop CMI to *M. lepraemurium*, that response was insufficient to restrain the replication of the mycobacteria.

These are several other interesting features of the *M. lepraemurium* counts. First, the bacilli multiplied more rapidly in the spleen after i.v. inoculation than in the LHF after local injection. Thus, the larger inoculum counts increased 100-fold in the spleen, but less than 10-fold in the LHF between weeks 0 and 8. The lower rate of replication in the footpad than the spleen has previously been attributed to the induction of CMI after LHF infection, but not after i.v. infection (13). That explanation is invalid because this phenomenon was observed in the nulnu as well as the nul+ mice. Second, the smaller inoculum multiplied more rapidly than the larger one, another phenomenon which, in other systems, is caused by the more rapid induction of CMI with larger inocula (9). Again, this effect of inoculum size occurred equally in the nul+ and nulnu mice. Third, the replication rate of *M. lepraemurium* in the spleen and LHF appeared slower during the first 4 weeks of infection than at some later intervals in nul+ and nulnu mice, which suggests bacterial lag rather than inhibition of bacteria by CMI.

A notable feature of the nude mouse experiments was the poor responsiveness of the nul+ mice. Thus, LHF swelling in response to 10^6 *M. lepraemurium* organisms did not occur until week 8, as compared to the more rapid responsiveness of other mouse strains whose feet became swollen by week 4 (13, 19). It is pertinent that the nul+ mice used in this study were derived from BALB/c mice, a strain which is notably susceptible and unresponsive to *M. lepraemurium* (1, 4, 6, 13). If, indeed, the nul+ mice were genetically unresponsive to *M. lepraemurium*, they might not develop immunity much more effectively than the nulnu mice.

This hypothesis was tested by comparing the responsiveness of nul+, CB6, and B6D2 mice to *M. lepraemurium* and *M. bovis* BCG. In earlier studies CB6 mice were shown to be genetically susceptible to *M. lepraemurium*, whereas the B6D2 mice were resistant (13), and both mouse strains respond strongly to *M. bovis* BCG. The footpad route of infection was employed to favor the induction of CMI, particularly in poorly responding mice. These experiments confirmed the slow onset of LHF swelling in nul+ mice in response to *M. lepraemurium*, with a concomitant delay in the induction of nonspecific resistance. However, once LHF swelling developed in nul+ mice it rapidly exceeded that in the other strains (Fig. 6), another indicator of high susceptibility (11). The footpad responses of CB6 and B6D2 mice to 10^6 *M. lepraemurium* organisms were closely similar (Fig. 6), but their responses to 10^8 bacilli revealed more rapid reactivity on the part of CB6 mice than on the part of B6D2 mice. So, paradoxically, if one accepts B6D2 mice as a paradigm of genetic resistance, then less resistant strains may respond either more briskly (CB6) or more slowly (nul+). The greater susceptibility of the nul+ and CB6 mice is evident from the numbers of *M. lepraemurium* organisms that were recovered from the LHF at 8 weeks.

The susceptibility of CB6, B6D2, and other mouse strains to *M. lepraemurium* is based on survival time after i.v. infection (13). It has been found that this distinction between susceptible and resistant strains is less apparent when survival time is measured after footpad infection (18, 24). However, it is abundantly clear from this study that high susceptibility to i.v. infection is associated with more rapid multiplication of *M. lepraemurium* in the foot after LHF inoculation. The responses to *M. bovis* BCG emphasize the specificity of genetic responsiveness to different mycobacterial pathogens. In contrast to their slow response to *M. lepraemurium*, nul+ mice respond very rapidly to *M. bovis* BCG; indeed, their hyperresponsiveness may signify increased susceptibility to *M. bovis* BCG, analogous to the response of CB6 mice to *M. lepraemurium*.

Because of their high susceptibility to *M. lepraemurium*, the nulnu and nul+ mice proved to be less useful for these studies than expected. Moreover, the relatively short survival of the nude mice, including uninfected controls, after introduction into the experiments rendered them unsatisfactory for the lengthy studies necessitated by *M. lepraemurium* infection. Consequently, further studies will be pursued with CB6 and B6D2 mice. The responsiveness of these mice to *M. lepraemurium* is established, and when they are depleted of T lymphocytes by adult thymectomy, lethal irradiation, and bone marrow reconstitution they survive for many months when housed with normal controls.

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LITERATURE CITED


