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Infection of the Congenitally Athymic Rat with *Mycobacterium leprae*[1]

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During the past decade a number of animal models have become available for the study of leprosy. These include intact and immunocompromised rodents and the armadillo (*Dasypus novemcinctus*). One advantage of small laboratory rodents is the large amount that is known about their physiologic, pathologic, and immunologic systems. This is particularly important if one wishes to study the role of host defense mechanisms which appear to be so important in the pathologic expression of clinical leprosy. In the initial studies of Shepard ([16]) using the foot pads of intact mice, inoculation of *Mycobacterium leprae* resulted in a limited, localized infection with a multiplication ceiling of approximately 10^6 bacilli per foot pad. Nonspecific suppression of T cell-mediated immunity of mice resulted in enhancement of *M. leprae* infection with approximately 10^7–10^8 organisms recoverable per foot pad, and intravenous inoculation resulted in a disseminated infection ([14]). A similar situation occurred in rats. For example, intravenous inoculation of neonatally thymectomized Lewis rats (NTLRs) resulted in a generalized infection with heavy bacillary infiltration of the cooler tissues, namely the snout, tail, ears, foot pads, and testes ([9]); whereas intravenous inoculation of intact rats resulted in little evidence of a generalized infection, although small numbers of organisms could sometimes be recovered from superficial tissues.

The congenitally athymic nude mouse also has been shown to develop disseminated infection following inoculation with *M. leprae* ([5]). However, nude mice must be maintained under very strict gnotobiotic conditions in order to survive long enough to develop a generalized infection. The congenitally athymic rat demonstrates a much greater ability to survive under normal laboratory conditions than the nude mouse ([8]). At the same time, it shares many of the properties associated with congenital absence of the thymus, including the acceptance of skin and tumor xenografts ([4, 6]), lack of response of splenic lymphocytes to T cell mitogens ([2, 18, 19]), and enhanced susceptibility to a number of infectious agents ([8]). We report here the characteristics of the infection resulting from the inoculation of *M. leprae* into athymic rats and compare this with the infection in intact rats and NTLRs.

MATERIALS AND METHODS

Animals. Normal and pregnant Wistar/Lewis rats were obtained from the Charles River Breeding Laboratories, Inc., Wilmington, Massachusetts, U.S.A. Neonatal thymectomy was carried out between 5 hr and 16 hr after birth using the method previously described ([9]). Athymic rats were obtained from our own colony, which was established from a small breeding nucleus of rats heterozygous for the *rnu* gene, obtained from the Laboratory Animal Centre, Carshalton, England. The *rnu* gene is being bred onto a Wistar/Lewis background by the cross-intercross system. The athymic rats used in the current study were derived from either the second or third cross with Wistar/Lewis rats.

Animal inoculation and assessment of bacillary growth. The strain of *M. leprae* used was originally isolated from a patient with leprosy by C. C. Shepard of the Centers for Disease Control, Atlanta, Georgia, U.S.A. It has been maintained in our laboratory for several years. Inoculation of *M. leprae,* har-

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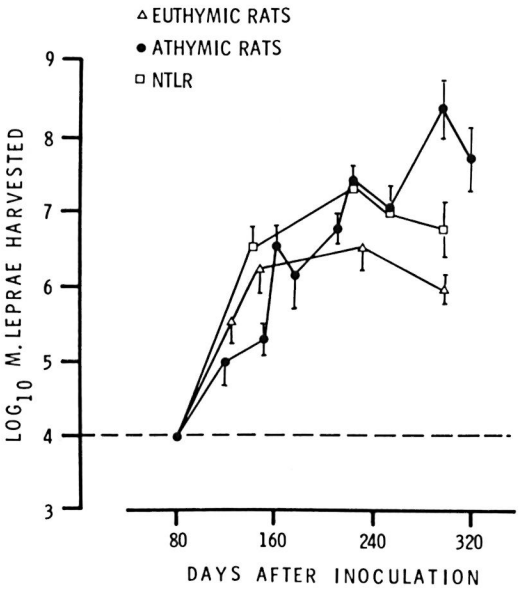

FIG. 1. Growth of *M. leprae* in the foot pads of athymic rats (●—●), euthymic rats (△—△), and NTLR (□—□) following inoculation into the foot pad of 5×10^3 organisms. Each point represents the mean of four foot pads (two rats). Error bars show standard error of the mean.

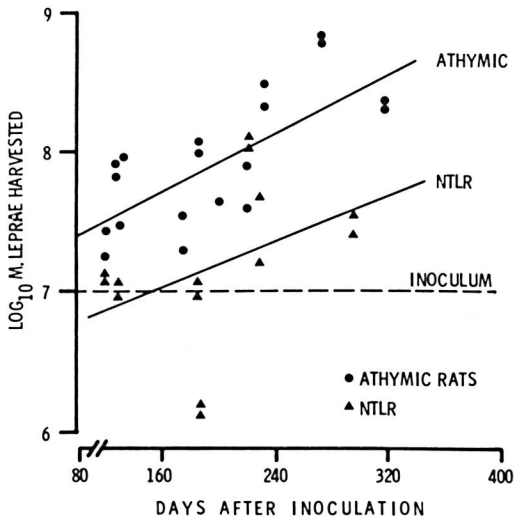

FIG. 2. Growth of *M. leprae* in the foot pads of athymic rats (●) and NTLRs (△) following inoculation of 10^7 organisms. Each point represents a single foot pad. The solid lines represent the lines of regression calculated for each group.

vesting of rat tissue, and counting of *M. leprae* were carried out by previously described methods ([17]). Foot pad and other tissues were processed and counted individually and, where appropriate, the mean value calculated. The number of tissues harvested is given in the figure legends.

Histology. Complete autopsies were performed on all animals. Tissues were fixed in 10% formalin and stained with hematoxylin and eosin (H & E) and Fite's stain ([10]). *M. leprae* were quantified by eye using the scale given in The Table. Semi-thin sections were prepared from formalin-fixed tissue embedded in glycomethacrylate, cut at 2.5 microns and similarly stained, except that for Fite's stain the time in carbol fuchsin was increased from 30 min to 45 min. For electron microscopy, one millimeter cubes of tissue were fixed in 2% gluteraldehyde, post-fixed in osmium tetroxide, and embedded in either Epon 812 or Araldite.

RESULTS

Foot pad inoculation with small numbers (5×10^3) of *M. leprae*. Groups of neonatally thymectomized Lewis rats, athymic, and intact control rats were inoculated in both hind foot pads with 5×10^3 *M. leprae*. Growth of the organism at this site was monitored for approximately 300 days. In control rats bacillary growth conformed to the classical growth pattern in normal rodents, reaching a ceiling of between 2×10^6 and 3×10^6 mycobacteria by 140 days post inoculation (Fig. 1). In comparison, growth of *M. leprae* was enhanced in NTLRs and reached a ceiling of 2×10^7. The growth curve of *M. leprae* in athymic rats was somewhat erratic, reaching a maximum of 2.6×10^8 per foot pad at 294 days, but falling at 317 days when the bacillary count was 5.4×10^7. Whether this represented a true limitation of growth or was simply a manifestation of the fairly large animal-to-animal variation is not clear. Dissemination of the infection to other superficial tissues was also assessed. Athymic rats harvested at 294 days and 317 days had tail counts of 3.2×10^5 and 2.6×10^5 *M. leprae*, respectively; whereas organisms could not be found in the front foot pads, ears, nose, or testes. In NTLRs, organisms were found only in the front and rear foot pads. In control rats there was no dissemination from the inoculation site.

Foot pad inoculation with large numbers (10^7) of *M. leprae*. Groups of adult athymic rats and NTLRs were inoculated in both

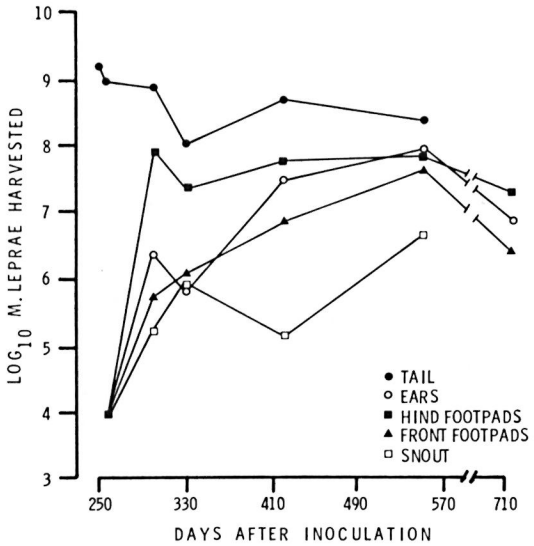

FIG. 3. Recovery of *M. leprae* from tail (●—●), ears (○—○), hind foot pads (■—■), front foot pads (▲—▲) and snout (□—□) of athymic rats following intravenous inoculation with 10^7 *M. leprae*. Each point represents a single sample.

hind foot pads with 10^7 *M. leprae* and bacillary growth monitored for 320 days. In all athymic rats harvested there was an increase in bacilli, with a maximum yield of 6.7×10^8 organisms at 272 days (Fig. 2). The counts in NTLRs were generally lower. The decrease in the number of organisms from the inoculum in one NTLR harvested at 187 days serves to emphasize the variability among these animals.

Dissemination of infection to superficial tissues occurred in both groups of rats from about nine months after inoculation. For example, one athymic rat harvested at 272 days had small numbers of bacilli in the tail (2.4×10^5), ears (6.0×10^4) and front foot pads (3.0×10^4), and an NTLR harvested at 295 days had organisms in the tail (7.8×10^6) and snout (1.5×10^4).

Intravenous inoculation of athymic rats with 10^7 *M. leprae*. Groups of athymic rats and NTLRs of both sexes aged 6–8 weeks were inoculated in the tail vein with 10^7 viable *M. leprae* and killed at intervals from 250–709 days, when bacillary counts as well as detailed pathologic examinations were performed.

During the course of the experiment the athymic rats gradually became cachectic, although there were no external signs of disease in the foot pads, snout, ears or tail. At autopsy the internal organs were grossly unremarkable, except for the lungs which showed a purulent bronchitis associated with areas of consolidation and collapse.

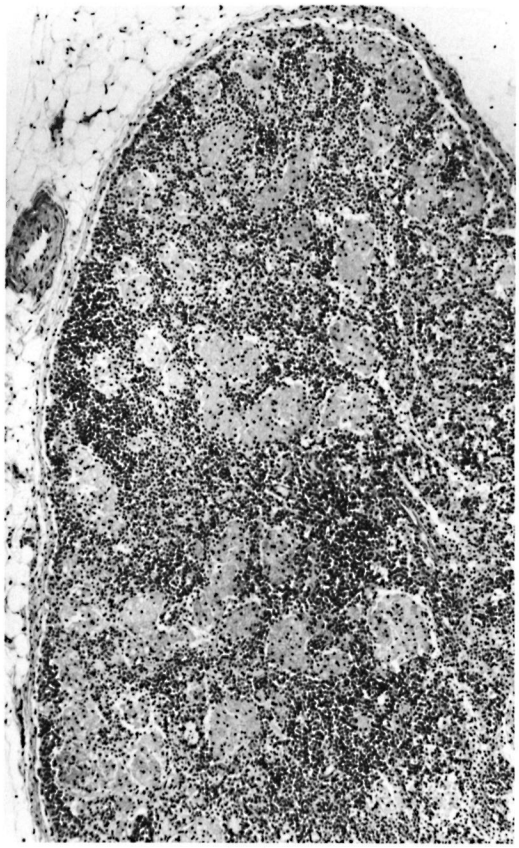

FIG. 4. Popliteal lymph node of athymic rat 12 months post intravenous inoculation with 10^7 *M. leprae* showing cortical depletion, absence of germinal centers and numerous granulomata (H & E ×80).

The result of the bacillary counts on athymic rats from 250–709 days post inoculation are given in Figure 3. At 250 days, counts in the tail had reached approximately 10^9 *M. leprae*. This was followed by an apparent decline in numbers to approximately 2×10^8 per tail by 530 days. By 300 days significant numbers of mycobacteria were recovered from all the tissues counted—the rear foot pads, front foot pads, ears, and snout, as well as the tail. However, there was a distinct tendency for the number of organisms at each site to plateau after one year at about 10^8, and in the

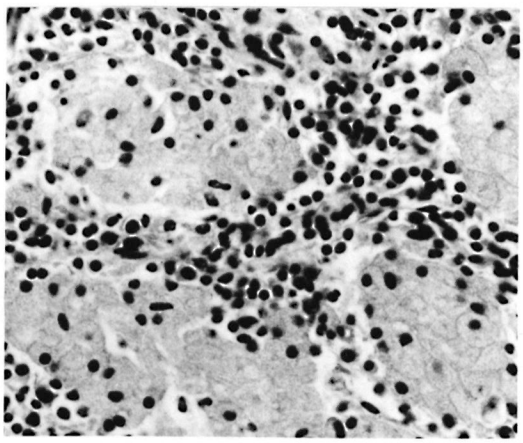

FIG. 5. Detail of granuloma from Figure 4. Note the vacuolated cytoplasm and small dark nuclei of the macrophages (H & E ×400).

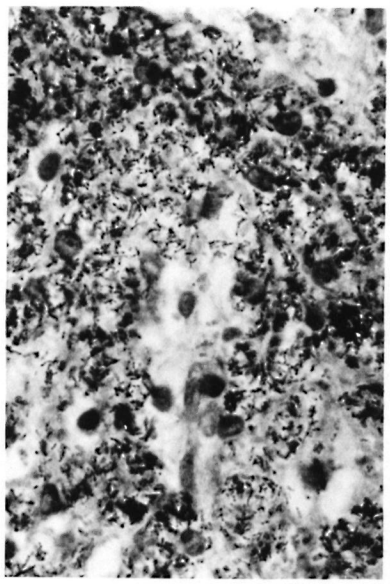

FIG. 6. Very large numbers of *M. leprae* in a granuloma in a popliteal lymph node (Fite ×1000).

last animal killed 703 days post inoculation the numbers of bacilli fell to only about 10^7.

These findings were correlated with the microscopic changes in the rats and the numbers of bacilli visualized in Fite-stained sections (The Table). The lymph nodes throughout the body showed reduction in the width of the cortex, with absent germinal centers and marked lymphocyte depletion of the paracortex. Mast cells and plasma cells were frequently present in the medullary sinuses. The most striking pathologic change was in the popliteal lymph nodes which contained variable numbers of small, sharply defined, non-caseating granulomata (Fig. 4). These were composed of large histiocytes with abundant, slightly vacuolated, eosinophilic cytoplasm and central, darkly staining, small nuclei (Fig. 5). These always contained large numbers (4+ to 5+) of *M. leprae* (Fig. 6); in contrast, only occasional organisms were present in macrophages elsewhere in the node. In addition, the popliteal nodes showed a variable degree of sinus histiocytosis which, in some cases, was marked. Lymph nodes elsewhere were less consistently involved. The axillary and inguinal nodes also contained variable numbers of granulomata associated with fairly large numbers of *M. leprae.* However, the more central nodes of the mediastinum were uninvolved in all rats examined. In the NTLRs, the lymph nodes showed a well-preserved cortical zone devoid of germinal centers. Present predominantly in the paracortical region were similar granulomata containing very large numbers of *M. leprae.* However, the numbers of granulomata were somewhat smaller than those seen in athymic rats.

The rear foot pads were involved in all cases but one. The inflammatory infiltrate was rather sparse and consisted of loose collections of histiocytes located just superficial to the plantar ligaments. Variable but, in general, rather large numbers of *M. leprae* were present in these cells. In one rat killed 15 months post inoculation the inflammatory response was more intense with large, irregular aggregates of histiocytes lying adjacent to the plantar facia. These infiltrated the muscle and along neurovascular bundles (Fig. 7). Very large numbers of *M. leprae* were present in these cells (Fig. 8). Small clusters of organisms were also present in striated muscle and a few organisms were present in small plantar nerves. In some animals there were, in addition, a few lymphocytes below the dermis. The front foot pads were only involved in animals killed at 21 months and 24 months post inoculation. The inflammatory changes and numbers of organisms present were considerably less than in the hind foot pad.

M. leprae were present in the tails of all animals and were most numerous (4+ to

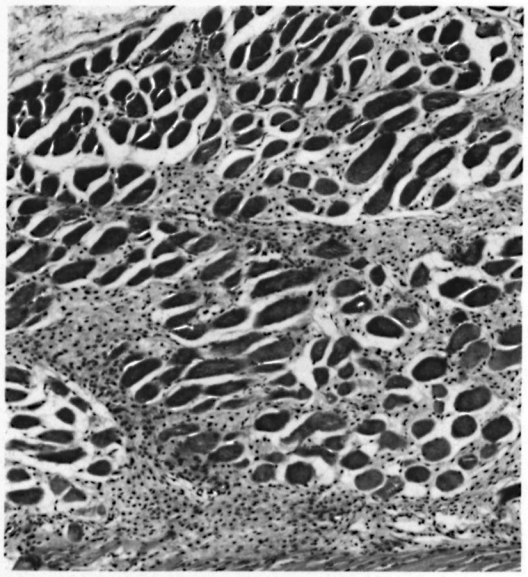

FIG. 7. Rear foot pad of an athymic rat 15 months post inoculation with *M. leprae* showing infiltration of nuclei by large numbers of foamy histiocytes (H & E ×80).

5+) in rats killed 12–15 months post inoculation. Bacilli were again present in large collections of histiocytes with foamy cytoplasm. In some animals these were located in the dermis but, more characteristically,

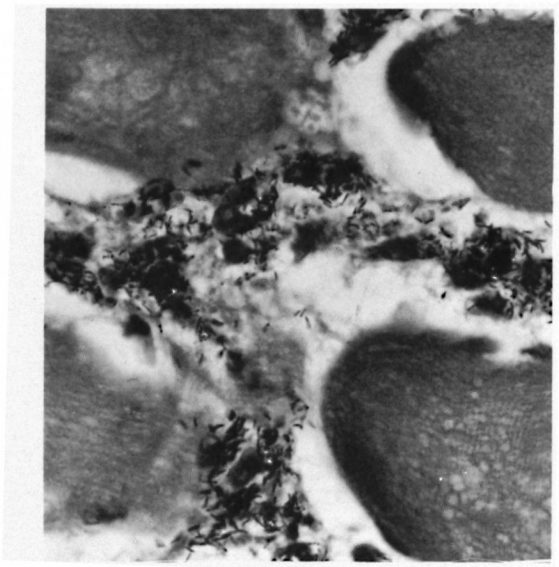

FIG. 8. The same specimen as in Figure 7, showing large numbers of *M. leprae* within histiocytes. Some of the mycobacteria appear to be in the same plane as the muscle fibers (Fite × 800).

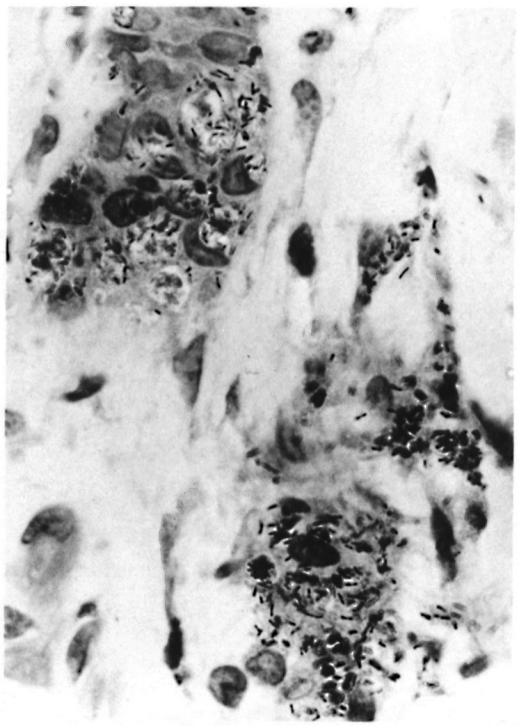

FIG. 9. Sections of tail of athymic rat inoculated two years previously, showing *M. leprae* within basal cells of hair shafts. (Glycomethacrylate embedded sections stained by a modified Fite stain ×880.)

they were located deep in the subcutis just external to the ligaments of the tail. Organisms were also located in fibroblast-like cells in the adventitia and walls of small vessels, and in the perineurium, as well as within small nerves. They frequently clustered around skin appendages and were present in the external layers of hair shafts (Fig. 9), as well as sebaceous cells and, rarely, epidermal cells. The NTLRs showed a similar infiltrate with, in one rat, small granulomata composed of foamy macrophages. The distribution of organisms was similar to that seen in the athymic animals.

The ear was likewise involved in all athymic rats. In general, there was a mild lymphohistiocytic infiltrate associated with the presence of very occasional clusters of organisms near the base of the pinna. In two animals killed 12 months and 24 months post inoculation, collections of foamy macrophages containing numerous mycobacteria were present in the dermis and between bundles of striated muscle at the base

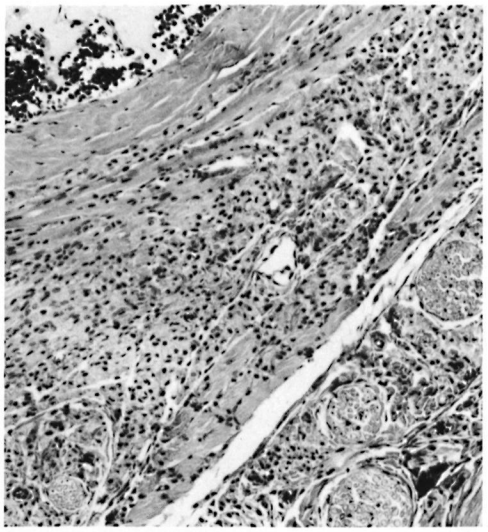

FIG. 10. Snout of athymic rat inoculated 15 months previously showing collections of foamy histiocytes lying beneath the skin and infiltrating along neurovascular bundles (lower right) (H & E ×80).

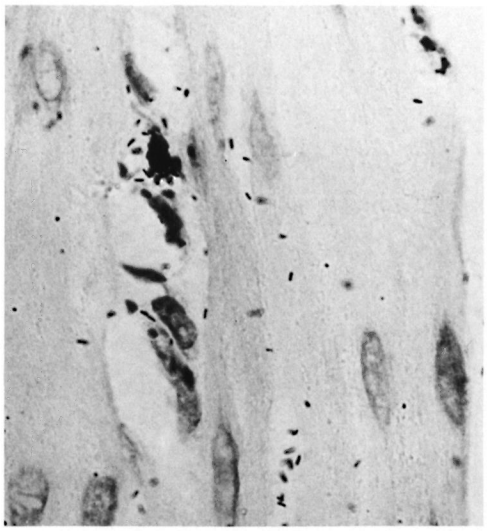

FIG. 11. Dartos muscle of scrotum showing mycobacteria lying in clusters adjacent to the nuclei and free in the sarcoplasm (Fite ×1000).

of the ear. In NTLRs very occasional collections of *M. leprae*-containing histiocytes were identified near the base of the ear. The changes in the snout were variable, but bacilli were present in all but one of the athymic rats and in all the NTLRs. In two athymic rats killed 12 months and 15 months post inoculation, there were granulomatous changes beneath the epidermis. These were particularly marked in the older animal, where they consisted of loose aggregates of histiocytes with granular cytoplasm that infiltrated around and between the skin appendages, muscle bundles, and along neurovascular bundles (Fig. 10). Large numbers of organisms were present within histiocytes and in more elongated cells, resembling fibroblasts. Bacilli were also present in perineural cells, small nerves, and muscle fibers. In the less severely involved animal the collections of histiocytes were smaller and the bacilli were generally confined to them. NTLRs also showed a rather wide spectrum of similar histologic changes. *M. leprae* were also observed in the liver, spleen, and bone marrow of athymic rats killed from 12 months to 15 months post inoculation, but not in rats harvested later. Although these organs appeared normal in hematoxylin and eosin-stained slides, Fite stains revealed small numbers of bacilli in cells of the mononuclear phagocyte series. In NTLRs, organisms were found in Kupffer cells in the liver throughout the experiment.

The only two male athymic rats available were killed at 15 months and 21 months post inoculation. Although the testes of the former were normal histologically and contained no organisms, large numbers of *M. leprae* were found in the scrotal skin. These were predominantly located in the dartos muscle, lying in clusters in clear vacuoles adjacent to the nucleus, but also apparently lying singly in the sarcoplasm (Fig. 11). In addition, they were present within histiocytes. They were, however, not seen in the bundles of striated muscle that were also present in the scrotum. The testes of the other male were atrophic, and mycobacteria were not identified in either the testes or scrotum. Routine sections of many other organs, including the sciatic nerve, diaphragm, thymic region, tongue, kidneys, and abdominal skin, revealed neither histologic abnormalities nor *M. leprae.*

Electron microscopy was carried out on the popliteal lymph nodes and tails of infected athymic rats. In the popliteal nodes very large numbers of organisms were present in macrophages that were characterized by the presence of numerous phagolysosomes (Fig. 12). Although some mycobac-

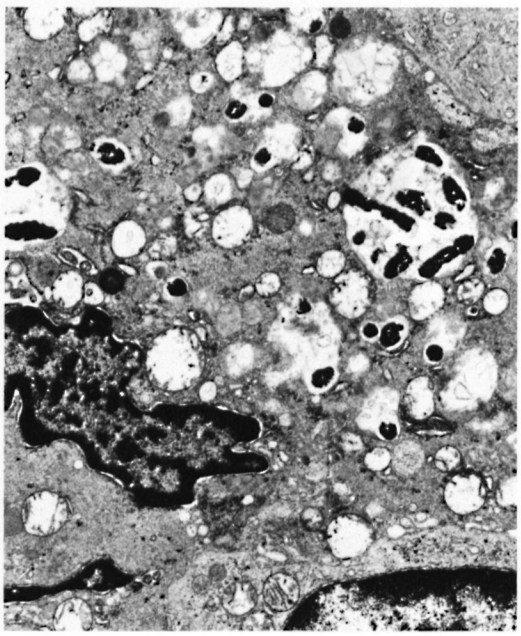

FIG. 12. Electron micrograph of part of histiocyte from a granuloma in a popliteal lymph node. Note the enormous number of phagolysosomes containing *M. leprae* (×8100).

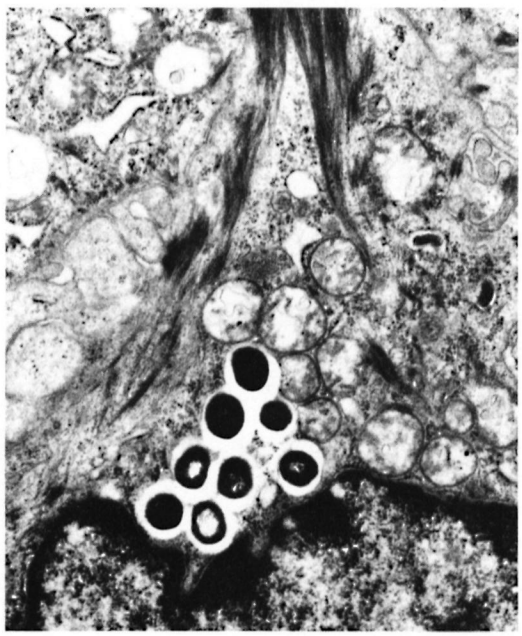

FIG. 13. Electron micrograph showing *M. leprae* lying within a squamous cell characteristized by desmosomes and abundant tonofilaments. This cell forms part of a hair sheath of the tail of an athymic rat inoculated 15 months earlier (×18,900).

teria appeared intact, the majority were degenerate. Nearly all were surrounded by a clear zone and contained within double membrane-bound vesicles. Sometimes the latter were sufficiently large to contain fragments of several organisms. In addition, the cytoplasm contained rather large numbers of lysosomes, small numbers of mitochondria, polyribosomes, and small amounts of smooth and rough endoplasmic reticulum. Cytoplasmic extensions were inconspicuous. The nuclei were round or oval with varying degrees of indentation. The cells thus corresponded to activated macrophages rather than epithelioid cells.

Electron microscopy of the tail revealed bacilli to be present not only in activated macrophages but also in squamous cells characterized by the presence of desmosomes and tonofilaments (Fig. 13). From the study of accompanying semi-thin sections, it was seen that these cells formed part of a hair sheath and that the bacteria-containing macrophages were deep in the dermis surrounded by collagen or muscle fibers. Bacilli were not seen in the samples from the rear foot pads examined by the electron microscope.

DISCUSSION

Our results demonstrate that athymic rats are more highly susceptible to infection with *M. leprae* than either NTLRs or euthymic rats. Inoculation of 5×10^3 bacilli into the foot pads resulted in an enhanced infection compared to NTLRs and intact control rats, and there was preliminary evidence of dissemination of the infection in both athymic rats and NTLRs. When an inoculum of 10^7 *M. leprae* per foot pad was used, a dose which is immunogenic in intact rats and mice, progressive bacillary growth occurred in both athymic rats and NTLRs, although the former contained higher numbers of organisms.

Following inoculation into the tail vein of athymic rats, bacterial counts were again higher than in NTLRs. This was particularly noticeable in the first year after inoculation. Subsequently a plateau of about 10^8 mycobacteria was reached in most of the cooler tissues. Fite stains showed that in animals killed 12 months to 15 months post inoculation, organisms were widely disseminated. They were appreciably more nu-

merous both in the superficial tissues and in the phagocytic cells of the liver and spleen than in NTLRs examined here or in our earlier studies ([7]). Subsequently they appeared to have been cleared from the liver and spleen, since they could not be found in animals killed 18 months or longer after inoculation.

Morphologically, the disease was qualitatively similar to that seen in NTLRs. There was, however, a good deal of variation in the intensity of the inflammatory response in individual animals. In the majority of athymic rats the inflammatory response was sparse and could be easily overlooked. However, in a few animals (notably, one killed at 15 months post inoculation) the response was striking, with large masses of foamy macrophages, reminiscent of lepromatous leprosy, infiltrating the snout and foot pads. When involvement of small nerves was present, it was found only in areas where there was a heavy infection. Granulomata were prominent in the lymph nodes of athymic rats but were also found in NTLRs, particularly in the popliteal nodes, a site that had not been routinely examined in our previous experiments ([7]). Surprisingly, the testes, which were extensively involved in NTLRs, were uninvolved, although in one animal the scrotal skin was heavily infected. Involvement of subcutaneous involuntary muscle of the scrotum was striking and while many mycobacteria were in histiocytes, others were quite clearly in smooth muscle cells. Electron microscopy showed the granulomata in the lymph nodes to be composed of macrophages which appeared to be activated, containing within phagolysosomes, numerous ingested mycobacteria, most of which were showing degenerative changes.

Although athymic rats develop disseminated infection with *M. leprae,* the evidence shown in Figure 3 suggests that they were capable of limiting the infection. Although we have no direct evidence for the mechanism involved, the morphologic findings suggest that macrophage activation could be an integral part of this. If this is so, then it raises the question of the mechanism of activation. Since the animals are athymic, a T cell-independent mechanism must be involved. Such a mechanism is known to exist in the nude mouse, which possesses non-specifically activated macrophages that are important in the control of acute intracellular infections ([3, 13]). These activated macrophages in nude mice appear to arise from direct stimulation by the microbial flora of the intestinal tract, and the activation is suppressed in mice which possess a T cell population ([15]). On the other hand, it is also possible that small numbers of cells bearing the surface antigens of T cells or T cell precursors may exist in the nude rat. Brideau, *et al.* ([1]) have shown that 9% and 10% of spleen cells from the athymic rat react with the monoclonal antibodies W3/25 and MRC.OX8, respectively. In athymic rats that develop lymphatic leukemia following the inoculation of the Friend-associated lymphatic leukemia virus, we have found cells in both the spleen and bone marrow that react with MRC.OX7 and W3/25 ([11]). However, it is not clear whether these cells are capable of functioning in the absence of intrathymic maturation.

The implication of the present study is that the athymic rats also possess activated macrophages, possibly to a greater extent than do athymic mice. It is interesting to note that Colston, *et al.* ([4]) found that xenogenic tumor cell lines often grew and then regressed in athymic rats. Dawson, *et al.* ([6]) had only limited success in attempting to transplant fresh human tumors, while Maruo, *et al.* ([12]) have found that acceptance of human tumor xenografts in athymic rats is age dependent, with older rats being significantly more resistant to tumor growth. Since activated macrophages are known to be involved in controlling tumor development, it is possible that macrophage activation levels increase with age in the athymic rats, as is known to occur in guinea pigs and in some strains of mice ([20, 21]). This would certainly explain the data shown in Figure 3 and The Table, in which there appears to be a slow decline in the level of infection after 12 months. What is not apparent from the present study is to what extent the *M. leprae* infection is involved in this apparent increase in macrophage activation.

SUMMARY

The susceptibility of congenitally athymic rats to *Mycobacterium leprae* infection has been investigated. Following inocula-

THE TABLE. *Comparison of distribution of* M. leprae *in the tissues of congenitally athymic rats and neonatally thymectomized irradiated Lewis rats following intravenous inoculation with 10^7 organisms.*

Tissue[a]	Time to sacrifice (months)								
	Congenitally athymic rats						Neonatally thymectomized rats		
	12	12	15	18	21	24	12	18	21
Rear foot pad	3[b]	0	5	2	5	4	2	4	N/A
Front foot pad	0	0	0	0	3	4	0	3	4
Snout	0	4	5	2	3	4	2	5	N/A
Ear	4	2	2	2	3	5	0	3	3
Tail	4	5	5	3	2	4	3	5	4
Spleen	1	2	2	0	0	0	0	2	3
Liver	2	2	2	0	0	0	1	1	2
Bone marrow	0	1	1	0	0	0	0	0	0
Popliteal lymph nodes	5	5	4	4	5	5	5	4	5
Scrotal skin	N/A	5	N/A	N/A	0	N/A	N/A	N/A	N/A

[a] Organisms were not identified in the other tissues examined.

[b] Because *M. leprae* were localized to only certain areas of the tissue it seemed preferable not to quantify them in terms of the number of fields examined. We, therefore, used the following scale:

1 plus = a single organism identified in the section.
2 plus = several organisms or clusters of organisms present.
3 plus = organisms easy to find.
4 and 5 plus = large and very large numbers present, respectively.

tion of small numbers of *M. leprae* (5×10^3) into the foot pad, the organisms replicated and attained a maximum of 2.6×10^8 per foot pad at 294 days; there was limited dissemination to the tail. In similarly inoculated neonatally thymectomized Lewis rats (NTLRs) a ceiling of 2×10^7 organisms was reached. When a larger inoculum (10^7) was given, the number of bacilli in athymic rat foot pads peaked at 6.7×10^8 and after approximately 240 days a plateau of between 2×10^8 and 6×10^8 per foot pad was reached. Dissemination to superficial tissues occurred approximately nine months after inoculation, when significant numbers of bacilli were recovered from the foot pads, ears, snout, and tail.

Following intravenous inoculation of 10^7 *M. leprae* into athymic rats, significant numbers of bacilli were recovered from the superficial tissues by 300 days post inoculation. The numbers of organisms reached a plateau of about 10^8 by one year. Autopsy of infected animals from 1–2 years after inoculation revealed no gross abnormalities except for a purulent bronchitis and bronchopneumonia. Although normal grossly, the ears, tail, snout and foot pads showed a varying degree of infiltration by histiocytes. In some this was almost imperceptible, in others there were large accumulations of foamy macrophages reminiscent of lepromatous leprosy. The numbers of mycobacteria present in Fite stains ranged from 2+ (several organisms or clusters of organisms) to 5+ (very numerous). The lymph nodes contained numerous non-caseating granulomata composed of activated macrophages which contained large (4+) or very large (5+) numbers of bacilli. Mycobacteria were present in the cells of the mononuclear-phagocyte series in the liver and spleen of animals killed 12–15 months post inoculation, but were absent from these cells in animals killed later. *M. leprae* were also numerous in the smooth muscle of the scrotum.

It is concluded that congenitally athymic rats are highly susceptible to *M. leprae* infection. Despite their lack of thymic-dependent T cell function, it appears that they possess the defense mechanism(s) capable of limiting the infection.

RESUMEN

Se investigó la susceptibilidad de las ratas congénitamente a ímicas a la infección por el *M. leprae*. Después de inocular números pequeños (5×10^3) de *M. leprae* en el cojinete plantar, los organismos se replicaron hasta alcanzar un máximo de 2.6×10^8 por co-

jinete plantar a los 294 días. Hubo limitada diseminación a la cola. En ratas Lewis timectomizadas neonatalmente e inoculadas de manera similar, se alcanzó un máximo de 2×10^7 microorganismos. Cuando se administró un inóculo mayor (10^7), el número de bacilos en los cojinetes plantares de las ratas atímicas alcanzó un pico de 6.7×10^8 y después de aproximadamente 240 días una meseta de 2×10^8 a 6×10^8 bacilos. La diseminación a los tejidos superficiales ocurrió aproximadamente 9 meses después de la inoculación, cuando se recuperaron números significativos de bacilos a partir de los cojinetes plantares, de las orejas, de la trompa y de la cola.

Trescientos días después de la inoculación intravenosa de 10^7 *M. leprae* en las ratas atímicas se pudieron recuperar números significativos de bacilos a partir de sus tejidos superficiales. Los números de organismos alcanzaron una meseta de aproximadamente 10^8 un año después de la inoculación. Las autopsias de los animales con una infección de uno a dos años no reveló grandes anormalidades excepto por una bronquitis purulenta y bronconeumonía. Aunque aparentemente normales, las orejas, la cola, la trompa y los cojinetes plantares mostraron un grado variable de infiltración histiocítica. En algunos, ésto fue casi imperceptible, en otros hubieron grandes acumulaciones de macrófagos espumosos reminiscentes de la lepra lepromatosa. Los números de micobacterias presentes en las preparaciones teñidas por Fite, oscilaron de +2 (varios organismos o grupos de organismos) a 5+ (muy numerosos). Los ganglios linfáticos contuvieron numerosos granulomas no caseosos, compuestos por macrófagos activados que contuvieron grandes (4+) o muy grandes (5+) números de bacilos. Las micobacterias estuvieron presentes en las células de la serie fagocítica-mononuclear en el hígado y en el bazo de los animales sacrificados 12 a 15 meses post-inoculación, pero no se encontraron en las células de los animales sacrificados después. También se encontraron numerosos *M. leprae* en el músculo liso del escroto.

Se concluyó que las ratas congénitamente atímicas son altamente susceptibles a la infección por *M. leprae.* No obstante su falta de función celular T dependiente del timo, parece que poseen otros mecanismos de defensa capaces de limitar la infección.

RÉSUMÉ

On a étudié la susceptibilité de rats congénitalement athymiques à l'infection par *M. leprae.* A la suite de l'inoculation d'une faible quantité de *M. leprae* (5×10^3) dans le coussinet plantaire. Les microorganismes se sont reproduits, atteignant un maximum de 2.6×10^8 par coussinet plantaire, après 294 jours. On a observé une dissémination limitée à la queue. Dans les rats de Lewis thymectomisés à la naissance (LTRSs), et inoculés de manière semblable, le nombre de microorganismes a atteint un plateau de 2×10^7. Lorsque l'on procédait à une inoculation de plus grandes quantités de bacilles (10^7) le nombre de microorganismes dans les coussinets plantaires du rat athymique montait jusqu'à 6.7×10^8, atteignant un plateau se situant entre 2×10^8 et 6×10^8 par coussinet plantaire après approximativement 40 jours. Une dissémination aux tissus superficiels est survenue environ neuf mois après l'inoculation; à ce moment, on pouvait recueillir des nombres significatifs de bacilles dans les coussinets plantaires, les oreilles, le museau et la queue.

A la suite d'une inoculation intra-veineuse de 10^7 de *M. leprae* chez des rats athymiques, des quantités significatives de bacilles ont été recueillies dans les tissus superficiels 300 jours après l'inoculation. Le nombre d'organismes atteint un plateau d'environ 10^8 après un an. L'autopsie des animaux infectés, pratiquée entre un et deux ans après l'inoculation, n'a pas révélé d'anomalies macroscopiques, à part une bronchite purulente et une broncho-pneumonie. Malgré leur aspect normal à première vue, les oreilles, la queue, le museau et les coussinets plantaires montraient différents degrés d'infiltration par des histiocytes. Chez certains animaux, cette infiltration était presque imperceptible, alors que chez d'autres, on pouvait observer une grande accumulation de macrophages spumeux, qui rappelait la lèpre lépromateuse. Le nombre de mycobactéries présentes à la suite d'une coloration par la méthode de Fite, s'étalaient de 2+ (plusieurs organismes ou amas d'organismes) à 5+ (très nombreux bacilles). Les ganglions lymphatiques contenaient de nombreux granulomes non-caséeux, composés de macrophages activés contenant une quantité élevée (4+) ou très élevée (5+) de bacilles. Des mycobactéries étaient présentes dans les cellules de la série phagocytaire mononucléaire, dans le foie et la rate d'animaux tués 12 à 15 mois après inoculation; par ailleurs, on ne trouvait pas de mycobactéries dans ces cellules chez les animaux sacrifiés plus tardivement. *M. leprae* a été également observé en grande quantité dans les muscles lisses du scrotum.

On en conclut que les rats congénitalement athymiques sont hautement susceptibles à l'infection par *M. leprae.* Malgré l'absence de la fonction des cellules-T dépendant du thymus, il apparaît que ces animaux possèdent des mécanismes de défense capables de limiter l'infection.

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