INTERNATIONAL JOURNAL OF LEPROSY

Volume 52, Number 4
Printed in the U.S.A.

Study of the Involvement of the Sciatic Nerve Following Inoculation with *M. leprae* and Other Mycobacteria in the Mouse Foot Pad[1]

A. N. Kamala, N. H. Antia, and V. P. Shetty[2]

Various studies have described nerve lesions in human leprosy at both the light microscopic and ultrastructural levels ([1, 2, 6, 7, 11, 14]). Weddell ([21]) demonstrated advanced lesions in the sciatic nerves of normal as well as immunosuppressed mice 20 months after inoculation of their foot pads with *Mycobacterium leprae*. Our previous studies, Antia ([3]), Mehta ([13]), and Shetty ([17–19]), have demonstrated typical lesions in the sciatic nerves of foot pad inoculated mice as early as the fourth post-inoculation month. These lesions were very similar to the changes demonstrated by us in the earliest stages of human leprosy as well as in contacts of leprosy patients. These early changes can only be observed at the ultrastructural level because they chiefly involve the unmyelinated fibers and their Schwann cells. They are observed as early as the fourth month and are well established by the eighth month after foot pad inoculation.

The type and sequence of nerve damage was similar in the early stages in both the tuberculoid and the lepromatous type of human disease ([17]), as well as in the sciatic nerve of the mouse ([19]), even though there was absence of acid-fast bacilli (AFB) in the nerves in both tuberculoid leprosy and the mouse model. Although the absence of bacilli in tuberculoid nerve lesions is well known, the diagnosis can be made confidently on the typical changes observed in the nerves.

It has been well established that the peripheral nerve and its Schwann cell are the target of *M. leprae* ([8, 10, 12]). The earliest lesions even in the biopsy of a skin patch are restricted to the dermal nerves, and any bacilli present are most likely to be detected in the nerve and its Schwann cell ([12]). So far as our present knowledge goes, the peripheral nerve is uniquely resistant to being affected by any other known bacterial infection except for the exotoxins of some organisms such as diphtheria. Lesions of the peripheral nerve have not been observed in any other mycobacterial disease, either in man or in animals. Even infections such as tuberculosis and *M. lepraemurium*, which are disseminated and involve a wide variety of tissues and organs, seem to spare the peripheral nerve in its entirety. All of this indicates that the involvement of the peripheral nerve in mycobacterial disease seems to be highly specific to *M. leprae*. Thus this may prove to be one of the most important criteria for characterization of this organism just as it is for the pathological confirmation of the diagnosis in this disease.

This study was undertaken to see if any of the other closely related mycobacteria inoculated into the mouse foot pad have the capacity to produce damage in its sciatic nerve and, if so, the nature of the lesion that is produced.

MATERIALS AND METHODS

The mycobacteria employed in this study were grouped as follows:

Group 1. *M. leprae* obtained from freshly harvested untreated lepromatous patients.

Group 2. Atypical mycobacteria that are known to occasionally produce human infection, *M. scrofulaceum* N-22, *M. intracellulare* N14061, and *M. avium* 844/M9 (obtained from Dr. J. Stanford, Middlesex Hospital, London, U.K.).

Group 3. Mycobacteria commonly used by investigators in leprosy, *M. vaccae* 859R (provided by Dr. Stanford), *M. phlei* NCTC

[1] Received for publication on 11 July 1983; accepted for publication in revised form on 23 May 1984.

[2] A. N. Kamala, M.Sc., Research Fellow, deceased; N. H. Antia, F.R.C.S., Trustee and Research Director; V. P. Shetty, Ph.D., Research Officer, The Foundation for Medical Research, 84-A, R. G. Thadani Marg, Worli, Bombay 400018, India.

TABLE 1. *Bacillary count following inoculation of mycobacteria into the mouse foot pad.*

Mycobacteria	Count per foot pad				
	4th mo.	6th mo.	8th mo.	16th mo.	20th mo.
M. leprae	$<1 \times 10^4$[a]	3.5×10^5	1.3×10^5	8.4×10^5	3.3×10^5
M. lepraemurium	7×10^7	3.2×10^8	8.9×10^8	ND[b]	ND
FMR 51 (4th passage)	2.4×10^6	7.3×10^6	1.02×10^6	ND	ND
FMR 75 (4th passage)	2.4×10^6	1.9×10^6	2.62×10^6	ND	ND
ICRC C-44	2.5×10^5	2.9×10^5	1.4×10^6	8×10^4	7.9×10^4
HI-75	1.08×10^6	2.9×10^5	$<1 \times 10^4$	$<1 \times 10^4$	$<1 \times 10^4$
M. scrofulaceum N-22	3.8×10^5	1.3×10^5	6×10^6	1×10^6	8×10^4
M. avium 844/M9	1.7×10^6	1.1×10^6	5.7×10^6	ND	ND
M. intracellulare N14061	1.3×10^5	$<1 \times 10^4$	$<1 \times 10^4$	$<1 \times 10^4$	$<1 \times 10^4$
M. vaccae 859R	1.3×10^5	$<1 \times 10^4$	$<1 \times 10^4$	$<1 \times 10^4$	$<1 \times 10^4$
M. smegmatis NCTC 10265	8.3×10^4	$<1 \times 10^4$	$<1 \times 10^4$	$<1 \times 10^4$	$<1 \times 10^4$
M. phlei NCTC 8151	3×10^4	$<1 \times 10^4$	$<1 \times 10^4$	$<1 \times 10^4$	$<1 \times 10^4$

[a] $<1 \times 10^4$ = no organisms seen in 100 fields.
[b] Foot pad harvest not done.

8151 and *M. smegmatis* NCTC 10265 (obtained from the Haffkine Institute, Bombay, India). *M. lepraemurium* were originally obtained from Dr. M. Nishiura, Kyoto, Japan, and are maintained in our laboratory by passaging in Swiss white mice. These cultivable mycobacteria were grown in Dubos medium and harvested during the log phase of growth.

Group 4. Mycobacteria which have been cultivated from biopsies of lepromatous leprosy patients, FMR strain 51 and FMR strain 75 both grown *in vitro* until the fourth passage and then inoculated into the mouse foot pad. In our laboratory these strains were grown in conditioned medium obtained from mouse dorsal root ganglion explant cultures, and they possibly belong to the *M. avium-intracellulare-scrofulaceum* (MAIS) group (avium-Battey Complex). The Skinsnes strain HI-75 (provided by Dr. Kato) belongs to the *M. scrofulaceum* complex ([20]). ICRC C-44 was obtained from Dr. C. V. Bapat of Cancer Research Institute. ICRC grown in Dubos medium also belongs to the avium-Battey Complex, in particular, *M. intracellulare*. Autoclaved *M. leprae* and normal saline were used as controls.

Random-bred, female Swiss white mice between 4 and 6 weeks of age were inoculated into each hind foot pad with 10^4 organisms in each group, 0.03 ml of saline being used in the control group. The mice were kept under identical conditions, and two mice from each group were killed at the end of the 4th, 6th, 8th, 16th, and 20th post-inoculation month. Both foot pads were harvested from each mouse and subjected to bacterial count as per the method of Rees ([15]). The sciatic nerves were fixed for electron microscopy by the technique which has been described previously ([16]). Briefly, the nerves were fixed in 2.5% glutaraldehyde, post-fixed in 1% osmium tetroxide, and embedded in epoxy resin. Semithin and ultrathin sections were cut, stained, and examined under the light microscope and 100S Jeol electron microscope, respectively. For light microscopy, the sciatic nerves were fixed in Zenker-formol overnight, washed in running water, dehydrated, and blocked in paraffin. Five μ sections were stained with Fite-Faraco and Triff to scan for the presence of AFB and 12 μ sections of the sciatic nerve were stained with Holm silver for the axon.

RESULTS

The number of acid-fast organisms harvested from the foot pads in each group at

TABLE 2. *Sciatic nerve involvement following inoculation of mycobacteria into the mouse foot pad.*

Mycobacteria	Electron microscopy				
	4th mo.	6th mo.	8th mo.	16th mo.	20th mo.
M. leprae	Unmyelinated fiber changes +[a]	Unmyelinated fiber changes +	Unmyelinated + Myelinated + (segmental)	Unmyelinated +++ Myelinated ++	Unmyelinated ++++ Myelinated ++++
M. lepraemurium	ND[b]	ND	NAD[c]	ND	ND
FMR 51 (4th passage)	Unmyelinated fiber +	Unmyelinated fiber +	Unmyelinated + Myelinated + (segmental)	ND	ND
FMR 75 (4th passage)	Unmyelinated fiber +	Unmyelinated fiber +	Unmyelinated + Myelinated + (segmental)	ND	ND
ICRC C-44	NAD	NAD	NAD	Unmyelinated ++ Myelinated ++	Unmyelinated +++ Myelinated +++
HI-75	NAD	NAD	NAD	NAD	NAD
M. scrofulaceum N-22	NAD	NAD	NAD	NAD	NAD
M. avium 844/M9	NAD	NAD	Unmyelinated + Myelinated + (axonal)	ND	ND
M. intracellulare N14061	NAD	NAD	NAD	NAD	NAD
M. vaccae 859R	NAD	NAD	NAD	NAD	NAD
M. smegmatis NCTC 10265	NAD	NAD	NAD	NAD	NAD
M. phlei NCTC 8151	NAD	NAD	NAD	NAD	NAD

[a] + = qualitative degree of nerve damage as observed at the ultrastructural level.
[b] Sciatic nerve biopsy not done.
[c] NAD = nothing abnormal detected in the nerve.

the 4th, 6th, 8th, 16th, and 20th months are shown in Table 1. In both the saline and autoclaved *M. leprae*-inoculated mice there was no growth of acid-fast organisms in the foot pads.

M. leprae obtained from fresh nodules of untreated lepromatous patients' biopsies showed the usual counts and growth curve with the logarithmic phase of growth up to the 6th and 8th post-inoculation months. FMR isolates 51 and 75 and *M. avium* showed persistent multiplication in the foot pads of the mice throughout the 4th, 6th, and 8th post-inoculation months, but the counts were at a higher level than those of *M. leprae.* HI-75 showed an initial growth of a million bacilli at the 4th month, then decreased at the 6th month; no organisms could be detected at the 8th, 16th, and 20th months. These results are comparable to those obtained by Stanford, *et al.* ([20]). *M. scrofulaceum* showed an increase from 3.8×10^5 at the 4th month to 6×10^6 at the 8th month and maintained a million bacilli at the 16th month. The count dropped to 8×10^4 at the 20th month; a slow grower rather like *M. leprae.* The ICRC bacillus had initial growth similar to *M. scrofulaceum* but showed a continuous level of growth with a first peak at the 8th month. *M. intracellulare, M. vaccae, M. smegmatis,* and *M. phlei* showed no growth in the foot pad.

Histopathological findings. At the light microscopic level there were no appreciable changes produced in any of the sciatic nerves, nor were any bacilli observed in any of the nerves. The foot pads were enlarged only in the *M. avium* and *M. lepraemurium* infections. Light microscopic preparations of the sciatic nerves of the *M. leprae*-inoculated mice showed no changes in the nerve morphology until the 16th month, at which time there was minimal Schwann cell proliferation. At the 20th month degenerative changes in the nerves were evident, but there was no infiltration or granuloma formation nor were any bacilli seen in the nerves that were examined. In *M. lepraemurium*-inoculated mice, the paraffin sections on light microscopic examination showed a normal complement of cells within the perineurial envelope. In Fite-Faraco- and Triff-stained sections, the myelin and axon appeared well preserved. Few bacilli were seen outside the perineurium, but none were seen within the nerve nor were there any infiltrating cells.

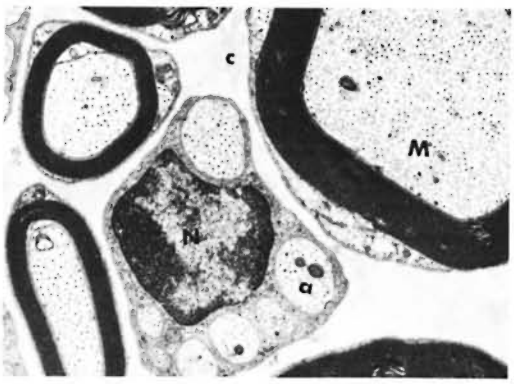

FIG. 1. Electron micrograph of an ultrathin section of normal sciatic nerve from the mouse used as control showing unmyelinated fiber (a) group as compact multiple axonal units supported by a single Schwann cell with a prominent nucleus (N); myelinated fiber (M) with intact neurotubules and neurofilaments, endoneurial collagen (C) seen (×20,000).

Ultrastructural findings (Table 2). The mouse sciatic nerve used as a control was taken at the same time as those of the experimental group of inoculated mice and showed normal features. The semithin sections consisted of one large, one small, and one very tiny funicle with a compact group of fibers.

At the electron microscopic level, the normal nerve (Fig. 1) revealed an unmyelinated fiber group as compact multiple axonal units surrounded by a single Schwann cell with a prominent nucleus. Myelinated fibers were well preserved with intact neurotubules and neurofilaments.

In *M. leprae*-infected mice, the semithin sections stained with toluidine blue did not show any changes until the 8th post-inoculation month, but at the 16th month demyelinating fibers and proliferation of the endoneurial blood vessels were seen. Observations at the 20th post-inoculation month indicated discrete areas of fiber loss; the sections also revealed degenerating and regenerating fibers.

At the ultrastructural level the sciatic nerve involvement was observed as early as the 4th post-inoculation month as reported earlier ([19]). The changes observed were those of hypertrophy of the Schwann cells of unmyelinated fiber groups and degeneration of

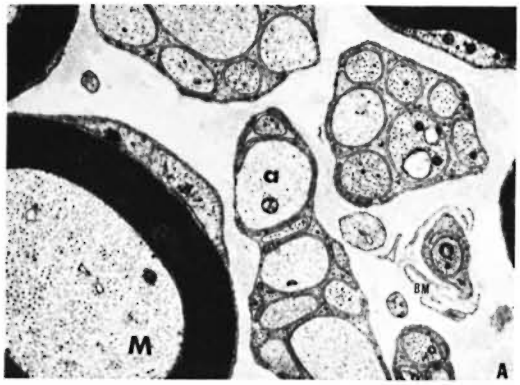

FIG. 2A. Electron micrograph of an ultrathin section of a sciatic nerve from a mouse inoculated into the foot pad with FMR 51 (*in vitro* grown, originally derived from human lepromatous tissue, Foundation for Medical Research) at the 8th post-inoculation month, showing degenerative changes among unmyelinated fiber groups. Swollen axon (a) has a foamy appearance of the axoplasm with disintegration of neurotubules and filaments. An incomplete Schwannian ensheathment (→) empty basal lamina (BM) around a well-preserved axon (a) probably indicates reinnervation following degeneration of an unmyelinated fiber unit; (M) = a well-preserved myelinated fiber (×16,000).

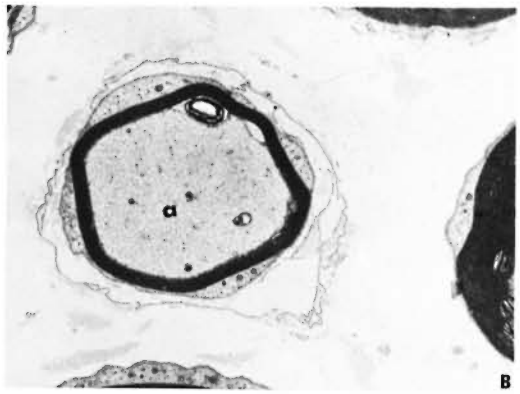

FIG. 2B. Another area from the same nerve showing a thinly myelinated axon (a) with an additional basement membrane tube, suggesting remyelination following segmental demyelination. The empty spaces proximal to the myelinated fibers indicate artefacts (×10,000).

their axons. This was followed by segmental demyelination of the small myelinated fibers as well as other changes, such as thickening of the basal lamina of the perineurium and an increase in endoneurial collagen comparable to early human nerve lesions as reported previously ([17]).

FMR isolates 51 and 75 showed changes similar to those of *M. leprae* (Figs. 2A and 2B). Unmyelinated fibers showed axonal swelling, disintegration of neurotubules and neurofilaments, and extremely small axonal sprouts surrounded by folded basement membrane tubes, indicating reinnervation following degeneration of an unmyelinated fiber unit. This involvement of the unmyelinated fiber at the 4th post-inoculation month persists and progresses with time. At the 8th post-inoculation month, both unmyelinated and myelinated fiber groups were involved. Figure 2B shows a thinly myelinated axon with an additional collapsed basement membrane tube, indicating remyelination following segmental demyelination.

The electron microscopic observations of the sciatic nerve of the mice inoculated with the ICRC bacillus showed qualitative nerve changes similar to those produced by *M. leprae,* but the nerve changes were quite evident only at the 16th post-inoculation month when all the nerves showed significant changes as compared to age-matched control nerves. Involvement of only a few unmyelinated fibers was seen at the 8th post-inoculation month, but this was not evident in all of the nerve samples. Changes were not consistent until the 16th post-inoculation month. On the other hand, in *M. leprae*-inoculated mice the sciatic nerve at the 4th post-inoculation month, in addition to unmyelinated fiber change, showed a significant thickening and a proliferation of Schwann cell processes forming concentric bands—Schwannian hypertrophy which was not observed with ICRC-infected mouse nerves. Blood vessels, although normal, had a dilated appearance. Perineurial cells also showed minimal vacuolation. Occasional axonal degeneration was also noted. The ultrastructural observations confirmed the semithin section findings of several either thinly myelinated fibers, or axons without any myelin, surrounded by a zone of Schwannian profiles.

Demyelination and remyelination were predominant among myelinated fibers. At two places Schwann cell cytoplasm showed two, transversely cut, dense bodies with a zone around them; those could have been ICRC bacilli.

The sciatic nerves of *M. lepraemurium*-infected mice did not show any fine struc-

tural changes and were comparable to the normal nerves used as controls; the myelinated and non-myelinated fiber groups were well preserved. There were no perineurial or blood vessel changes.

Semithin sections of the *M. avium*-inoculated mouse sciatic nerve showed degenerated axons with myelin debris in the surrounding region. At the ultrastructural level this nerve showed no abnormality at the 4th month, but at the 8th post-inoculation month axonal type degeneration in the sciatic nerve was apparent. There were small isolated areas of fiber loss with regenerating units, as shown in Figure 3, represented by multiple, small axonal sprouts around a freshly myelinated axon. These appear after the axon is degenerated, nascent folded basement membranes almost demarcating the regenerating units. Myelin debris is seen within macrophage-like cells. The blood vessels were not dilated, but there was a prominence of endothelial cells; the perineurium looked normal. These changes seen with *M. avium* infection in the sciatic nerve are unlike those normally observed in leprous nerves where there is progressive segmental demyelination of fibers with an intact axon, only Schwann cell involvement and no active degeneration. None of the other mycobacteria showed any detectable changes in the sciatic nerve of the inoculated mice, even at the ultrastructural level.

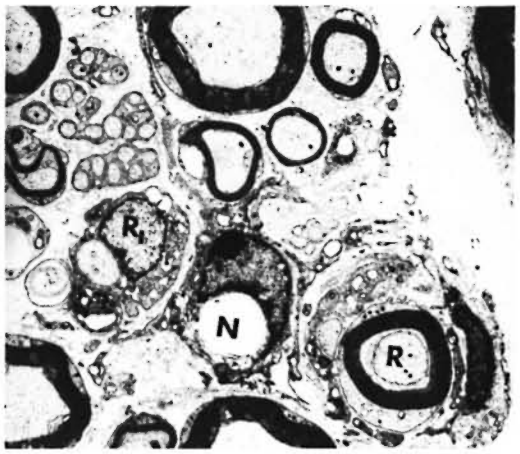

FIG. 3. Electron micrograph of an ultrathin section of sciatic nerve from a mouse inoculated into the foot pad with *M. avium* at the 8th post-inoculation month, showing two regenerating units (R) and (R_1) with additional folds of basal lamina and cell processes. A nucleated cell seen with a large phagocytic vacuole (N) is probably a macrophage. Some of the unmyelinated axons also show degenerative changes (×3828).

DISCUSSION

The involvement of the peripheral nerve is the most constant feature of leprosy and seems to be very specific to infection by *M. leprae.* There is no information available regarding peripheral nerve damage caused by any other bacteria, including other mycobacteria. If this is true, then the presence of nerve damage may prove to be one of the most important criteria for characterizing any acid-fast organism which claims to be *M. leprae.*

In view of the controversy over the characterization of organisms cultivated from leprosy patients, it was decided to use the mouse sciatic nerve model to see if mycobacteria other than *M. leprae* could produce the typical damage seen at the ultrastructural level in this model.

The 12 mycobacteria selected for this study consisted of both fast and slow growers. Group 2 consisted of known human pathogens. Group 3 consisted of a nonhuman pathogen MLM, and other organisms which have frequently been used by investigators in the field of leprosy. Group 4 consisted of organisms originally derived from human lepromatous tissue and later cultivated in artificial media.

A standard inoculum of 10^4 organisms was inoculated into the mouse foot pad as used for *M. leprae* and the growth curve, if any, was recorded. Growth curves in the foot pad were fairly similar to those recorded for these mycobacteria by other observers ([5, 9, 20]).

No clinical or histological nerve damage has been reported by any of these authors, and no ultrastructural studies have been undertaken by them. The only exception has been Wiersema, *et al.* ([22]), who described infiltration of MLM and macrophages containing acid-fast bacilli in the auricular and sciatic nerves of mice observed from the seventh month onward after inoculation. Our study did not reveal any infiltration in the sciatic nerve on light or electron microscopy, even when there was extensive spread of the organisms in almost all of the other tissues of the body.

Binford ([4]) reported the failure of the NQ

mycobacterium to invade nerves, although they frequently were completely surrounded by histiocytic granulomatous tissue containing numerous bacilli. Only two examples of intraneural growth have been seen in the numerous transmission experiments.

Of the mycobacteria used in this study, *M. intracellulare, M. vaccae, M. smegmatis,* and *M. phlei* showed minor growth at the fourth month, disappearing at the sixth month; none of these organisms showed nerve changes.

M. lepraemurium-inoculated mice did not reveal any nerve damage at the eighth month, even when the foot pad counts were high. FMR 51 and 75 cultured isolates and *M. avium* showed a level of growth of about one log higher than that of *M. leprae.* All of these organisms showed nerve damage at the ultrastructural level. While the damage caused by the FMR strains was typical of that caused by *M. leprae,* that is, involvement of the Schwann cells, damage due to *M. avium* was an entirely different axonal type of damage. Studies on these organisms could not be continued because of the death of the mice. The mortality of *M. avium*-inoculated mice after 15 months has been reported by Collins, *et al.* ([5]). *M. scrofulaceum* and the ICRC bacillus showed persistence of organisms similar to *M. leprae,* but *M. scrofulaceum* did not show any nerve damage even after this prolonged period of time.

The ICRC bacillus showed evidence of nerve lesions only from the 16th post-inoculation month, and these lesions were similar to those observed in the *M. leprae*-infected animals at the 16th post-inoculation month. Possibly the reason for the delay in the onset of nerve damage and for the less vigorous response from Schwann cells could be that the ICRC bacilli might have lost pathogenicity during *in vitro* cultivation. To regain pathogenicity *in vivo* might take time; hence the delay.

M. leprae inoculated into the foot pad of the mouse produce certain typical changes in the sciatic nerve. These have not been demonstrated in any of the other human pathogenic or nonpathogenic mycobacteria listed in this study. Three strains of mycobacteria cultivated from leprosy patients' biopsies, FMR strains 51 and 75 and ICRC bacilli, showed nerve changes similar to that of *M. leprae.* Thus, the typical sequential changes seen in the sciatic nerve of the mouse model may be used as one of the criteria for characterizing cultivable mycobacteria which are claimed to have the characteristics of *M. leprae.*

SUMMARY

In order to determine whether *Mycobacterium leprae* alone produce the typical damage in the sciatic nerves of foot pad inoculated mice as demonstrated earlier, a comparative study was undertaken using various other mycobacteria inoculated into the hind foot pads of normal Swiss white mice. The findings indicate that FMR isolates No. 51 and No. 75 and *M. avium* showed multiplication in the foot pads of the mice throughout the 4th, 6th or 8th post-inoculation months and these infections were associated with neural changes in the sciatic nerves. The type of nerve involvement in the case of *M. avium* differs from *M. leprae* in being predominantly an axonal degeneration at the 8th post-inoculation month, that is, degeneration of the complete axon and myelin debris remnants; whereas in *M. leprae* infection, where segmental demyelination predominates, the axons are intact and it is the Schwann cell that is affected. The neural changes in the case of FMR isolates No. 51 and No. 75 were similar to those seen in mice inoculated with *M. leprae* obtained directly from human biopsies.

Other mycobacteria, HI-75 (Skinsnes) and *M. scrofulaceum,* showed growth in the foot pad initially which persisted in the case of *M. scrofulaceum* until the 20th post-inoculation month, but no ultrastructural changes were observed in the sciatic nerves of these mice.

In ICRC-inoculated mice, nerve lesions were seen much later (at the 16th post-inoculation month) and the changes were similar to those seen with *M. leprae.*

M. vaccae, M. smegmatis, M. phlei, and *M. intracellulare* showed almost no growth in the foot pads of the mice, and there were no detectable changes in the sciatic nerves. *M. lepraemurium* showed growth in the foot pad but no lesions were seen in the sciatic nerve.

The study reveals that *M. leprae* inoculated into the foot pad of the mouse produce

certain typical changes in the sciatic nerve which have not been demonstrated with any of the other human pathogenic or non-pathogenic mycobacteria used in this study. Only three strains of mycobacteria cultivated from biopsies of leprosy patients, namely, FMR strains No. 51 and No. 75 and ICRC, demonstrated changes typical of those produced by *M. leprae.* In view of the fact that the peripheral nerve and its Schwann cells is the target of *M. leprae,* the typical changes seen in the sciatic nerve of the mouse model may be used as one of the criteria for characterizing any cultivable organism claiming to be *M. leprae.*

RESUMEN

Para determinar si el *Mycobacterium leprae* produce, per se, el daño típico en los nervios sciáticos de ratones inoculados en el cojinete plantar, se hizo un estudio comparativo usando *M. leprae* y varias otras micobacterias inoculadas en los cojinetes plantares posteriores de ratones Swiss-White. Los hallazgos indican que las muestras No. 51 y No. 75, y el *M. avium,* mostraron multiplicación en los cojinetes plantares del ratón hasta el 4°, 6°, u 8° mes post-inoculación y que estas infecciones estuvieron asociadas con cambios neurales en los nervios sciáticos. El tipo de daño del nervio en el caso del *M. avium* difiere del daño causado por el *M. leprae* en que en el primer caso predomina una degeneración axonal hacia el 8° mes post-inoculación, esto es, una degeneración del axón completo y de los restos remanentes de mielina, en tanto que en la infección por *M. leprae,* donde predomina la desmielinización segmental, los axones estan intactos y las células de Schwann son las afectadas. Los cambios neurales en los "aislados" No. 51 y No. 75, fueron similares a aquellos observados en ratones inoculados con *M. leprae* obtenidos directamente de biopsias humanas.

Otras micobacterias, HI-75 (Skinsnes) y *M. scrofulaceum,* mostraron un crecimiento inicial en el cojinete plantar que persistió, en el caso de *M. scrofulaceum,* hasta el mes 20 post-inoculación pero no se observaron cambios ultraestructurales en los nervios sciáticos de estos ratones.

En los ratones inoculados con la cepa ICRC, las lesiones en nervios se observaron mucho después (hasta el mes 16 post-inoculación) y los cambios fueron similares a los observados con *M. leprae.*

M. vaccae, M. smegmatis, M. phlei, y *M. intracellulare,* casi no mostraron crecimiento en los cojinetes plantares del ratón, y no hubieron cambios detectables en los nervios sciáticos. *M. lepraemurium* creció en el cojinete plantar del ratón pero no se observaron lesiones en el nervio sciático.

El estudio revela que el *M. leprae* inoculado en el cojinete plantar del ratón produjo ciertos cambios típicos en el nervio sciático que no se encontraron con ninguna de las otras micobacterias patogénicas o no patogénicas para el humano usadas en este estudio. Sólo tres cepas de micobacterias cultivadas de biopsias de pacientes con lepra (cepas No. 51 y No. 75 y la ICRC) demostraron cambios típicos similares a los producidos por *M. leprae.* En vista de que el nervio periférico y sus células de Schwann son el blanco del *M. leprae,* los cambios típicos observados en el nervio sciático del ratón, pueden ser usados como uno de los criterios para caracterizar a cualquier organismo cultivable que se sospeche sea *M. leprae.*

RÉSUMÉ

Afin de déterminer si *Mycobacterium leprae* seul peut entraîner des lésions typiques du nerf sciatique chez des souris inoculées au niveau du coussinet plantaire, ainsi que cela a été trouvé précédemment, on a procédé à une étude comparative des effets produits par diverses autres mycobactéries inoculées dans les coussinets plantaires des pattes arrières chez des souris blanches Swiss normales. Les observations recueillies montrent que les souches FMR 51 et 75, de même que *M. avium,* se multiplient dans les coussinets plantaires de la souris quatre, six, ou huit mois après l'inoculation; ces infections étaient associées avec des modifications des nerfs au niveau du sciatique. Le type de lésion nerveuse observé après inoculation de *M. avium* était différent de celui produit par *M. leprae,* car il consistait surtout en une dégénérescence des axones au huitième mois après l'inoculation. Cette dégénérescence portait sur l'entièreté de l'axone et des débris de myéline qui persistaient. Par contre, lors de l'infection par *M. leprae,* c'est la démyélinisation segmentaire qui prédominait, les cellules de Schwann étant atteintes alors que les axones demeuraient intactes. Dans le cas des souches FMR 51 et 75, les modifications des nerfs étaient semblables à celles observées chez des souris inoculées par des bacilles *M. leprae* recueillis directement de biopsie humaine.

D'autres mycobactéries, le HI-75 de Skinsnes et *M. scrofulaceum,* ont présenté une croissance initiale dans le coussinet plantaire qui, dans le cas de *M. scrofulaceum,* a continué jusqu'au 20ème mois après l'inoculation; aucune modification de l'ultrastructure n'a cependant été observée au niveau du nerf sciatique chez ces souris.

Chez les souris ICRC-inoculées, les lésions nerveuses étaient observées beaucoup plus tard, au 16ème mois après l'inoculation, et les modifications étaient semblables à celles trouvées après inoculation par *M. leprae.*

D'autres bacilles, à savoir *M. vaccae, M. smegmatis, M. phlei* et *M. intracellulare,* n'ont présenté quasiment aucune croissance dans les coussinets plantaires de la souris; aucune modification décelable n'a été relevée au niveau du nerf sciatique. *M. lepraemurium* a présenté une croissance dans le coussinet plantaire, sans qu'aucune lésion puisse être mise en évidence dans le nerf sciatique.

Ces études démontrent que *M. leprae* inoculé dans le coussinet plantaire de la souris entraîne certaines modifications typiques au niveau du nerf sciatique, qui n'ont été observées avec aucune autre des mycobactéries humaines pathogènes ou non pathogènes utilisées dans cette étude. Trois souches seulement de mycobactéries cultivées à partir de biopsies de malades de la lèpre, les souches FMR 51 et 75 et le bacille ICRC, ont entraîné des modifications typiques semblables à celles produites par *M. leprae*. Comme le nerf périphérique et les cellules de Schwann qui l'entourent constituent la cible d'élection de *M. leprae*, les modifications typiques mises en évidence chez la souris au niveau du nerf sciatique peuvent être utilisées comme un critère, parmi d'autres, pour identifier les organismes cultivables dont on réclame l'identité avec *M. leprae*.

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