Lepr. Rev (1969) **40**, 9-16

Leprous Myositis—A Histopathological and Electron-microscopic Study

C. K. JOB, A. B. A. KARAT, S. KARAT AND M. MATHAN

Schieffelin Leprosy Research Sanatorium, Karigiri

and

Christian Medical College and Hospital, Vellore, S. India

Microscopic and electron-microscopic examination of biopsy specimens of striated muscle and of the dartos tunica of the scrotum obtained from patients with lepromatous leprosy showed that the changes in leprous lesions of muscle occurred in 3 stages, viz. invasion by and proliferation of *Myco. leprae*, followed by degeneration of the muscle fibres and infiltration by leukocytes, and finally by replacement of the muscle fibres by fibrous tissue. Several lepromatous granulomata were observed in areas around blood vessels in the perimysium.

Mycobacterium leprae was cultured for the first time, though with limited multiplication, by Shepard (1962) in the footpads of mice. Later, Palmer *et al.* (1965) localized the exact site of multiplication of the organism in the striated muscles of the footpad. It is said (Rees and Weddell, 1968) that within a few hours of injecting the bacilli into the footpads they can be found in the muscle cells. However, in the human patient leprous lesions in the muscle are reported to be infrequent. As far as we know, in the literature there are only 3 papers, 2 describing leprous lesions of striated muscle (Ishihara, 1959; Convit *et al.*, 1960) and one demonstrating the infection in the dartos tunica (Harman, 1968).

In this present paper we describe the histopathological and the electron-microscopic appearance of leprous myositis in both striated and smooth muscles, and point out that the infection of superficially situated smooth-muscle cells is not an uncommon finding in lepromatous leprosy.

MATERIALS AND METHODS

In all, 8 muscle biopsy specimens of lesions from patients with lepromatous leprosy were available for study. Of these, 6 were from the striated muscles in different parts of the body and 2 from the scrotum, including the dartos tunica, the smooth muscle bundles situated just beneath the scrotal skin.

All the biopsy samples were fixed in 10% formalin, processed, blocked in paraffin wax, sectioned at 6μ thickness, and stained with the following stains: haematoxylin and eosin, acid-fast stain, Gömöri methanamine silver (GMS) stain, and Masson's trichrome stain. Two of the specimens were stained with Van-Gieson's, periodic-acid-Schiff (PAS), and Bodian stains. Since there had been no previous intention to include an electron-microscopic study only neutral formalin-fixed tissues were available in 2 cases with infection of striated muscle. However, one of the scrotal biopsy samples was fixed in paraformaldehyde. These biopsy specimens were later treated with osmium tetroxide, embedded in araldite, cut in a Cambridge Huxley microtome, and examined under a Philips EM 200 electron-microscope.

FINDINGS

The changes occurring in leprous lesions of the striated muscles were seen in 3 different stages. First, an initial stage of invasion and proliferation of the *Myco. leprae* inside muscle fibres and tissue histiocytes. Subsequently, the muscle fibres degenerated and were infiltrated by polymorphonuclear leukocytes, lymphocytes, and macrophages; the bacilli then became frag-

mented and granular. Finally, the destroyed muscle fibres were replaced by fibrous tissue, the macrophages became vacuolated, and the bacilli disappeared completely.

The active lesions present in 2 of the 6 cases studied, showed slight swelling and thickening of the endomysium. Several lepromatous granulomata (Fig. 1) were present in focal areas, around blood vessels in the perimysium. Adjacent muscle fibres were mostly normal, with longitudinal and cross striations well preserved. However, some of the fibres showed marked vacuolation, loss of striations and even necrosis. PAS staining was negative, indicating absence of glycogen. In some areas the sarcolemmal nuclei were clumped together. A few lymphocytes were also seen scattered diffusely in the muscle tissue. Acid-fast stain and GMS stain revealed numerous bacilli inside macrophages and muscle cells (Fig. 2). A few of these were

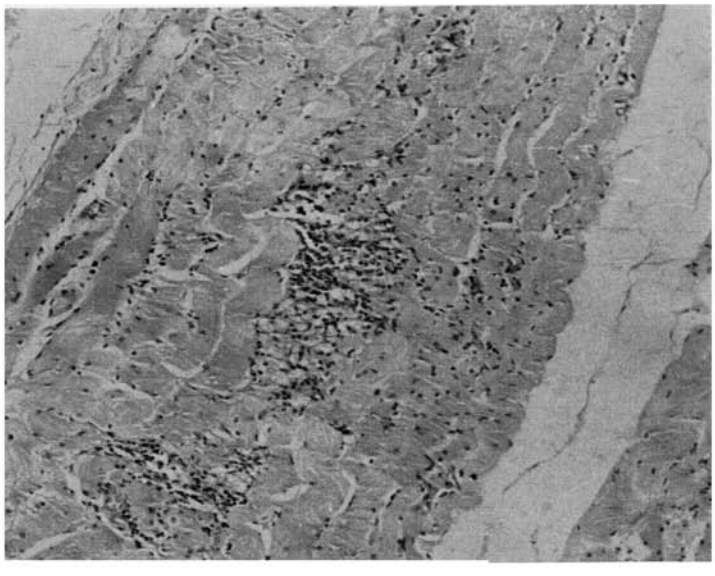

FIG. 1
Photomicrograph showing striated muscle with a lepromatous granuloma composed of foamy macrophages and round cells (H & E ×250).

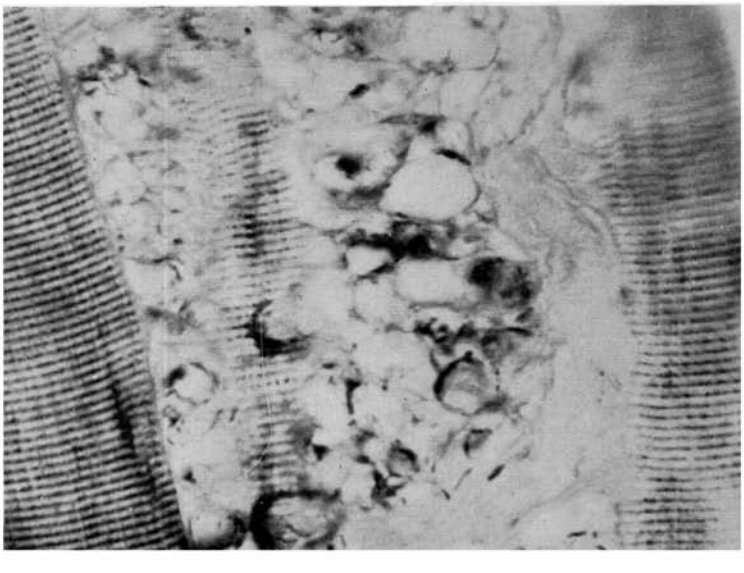

FIG. 2
Photomicrograph to show striated muscle fibres replaced by foamy macrophages containing *Myco. leprae*. The bacilli were found inside muscle cells also (GMS ×2000).

rods and a large number were granular organisms.

As the muscle fibres degenerated there was clumping of sarcolemmal nuclei. In some areas these were arranged in rows one behind the other, but in other areas 5 or 6 nuclei were seen in aggregation. The inflammatory granuloma at this stage consisted of a large number of macrophages, plasma cells, lymphocytes, and polymorphonuclear leukocytes the majority of which were eosinophils. The macrophages showed marked vacuolation (Fig. 3). Acid-fast stain and GMS stain revealed only a few organisms, most if not all of which were fragmented and granular. Several microscopical fields had to be searched before granules of acid-fast bacilli could be detected inside macrophages.

During the healing stage the necrosed muscle fibres were gradually replaced by fibrous tissue; the remaining muscle fibres showed well marked

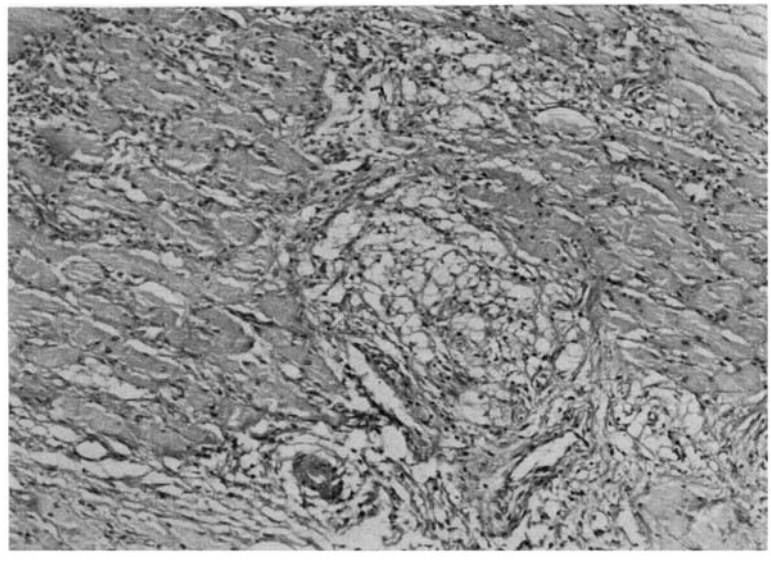

FIG. 3
Photomicrograph of a healing lepromatous granuloma in the striated muscles. Note the collection of vacuolated macrophages and the diffuse fibrosis of the muscle (H & E ×250).

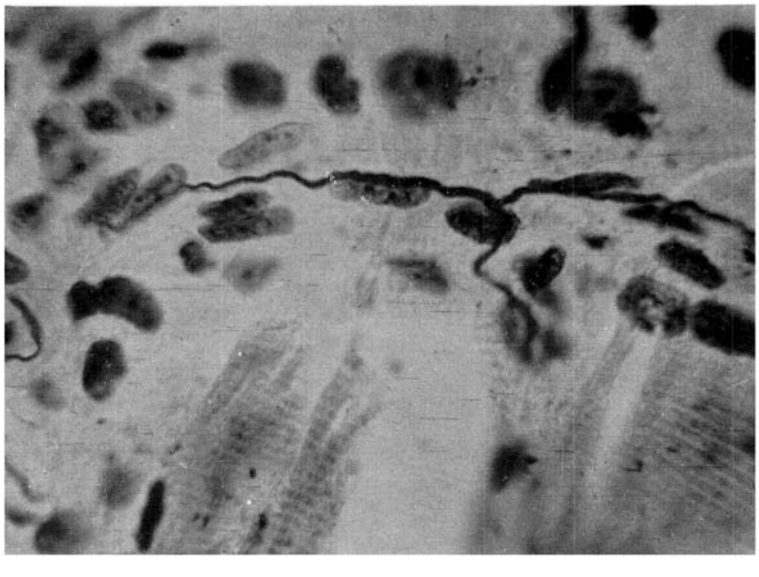

FIG. 4
Photomicrograph showing nerve fibres supplying muscle bundles infiltrated with numerous inflammatory cells (Bodian ×2000).

striation. In the longitudinal section, muscle fibres were interrupted at intervals by focal areas of fibrous tissue, suggesting that the necrosis of muscle fibres perhaps had been focal and patchy. There was diffuse infiltration of the fibrous tissue by lymphocytes, plasma cells, and a few polymorphonuclear leukocytes. The macrophage collections were still present but were markedly vacuolated; acid-fast stain showed no bacilli in them.

Bodian stain showed that the nerve fibres supplying the muscle cells were infiltrated with inflammatory cells (Fig. 4).

The 2 biopsy specimens from the dartos muscle of the scrotum were from active lesions. They showed that the muscle bundles were oedematous and swollen and some were obviously vacuolated. There were focal collections of macrophages, lymphocytes, and plasma cells around the muscle bundles (Fig. 5). Some of

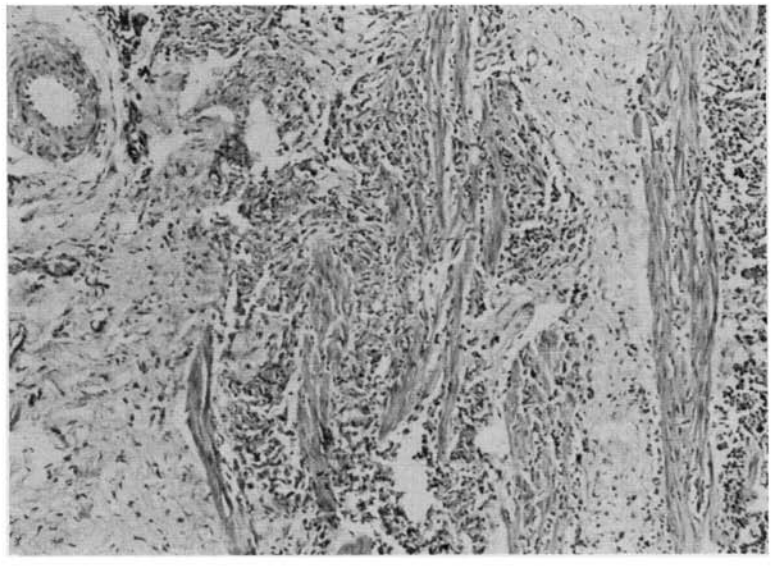

FIG. 5
Photomicrograph to show smooth-muscle bundles from dartos tunica infiltrated by lymphocytes and plasma cells (H & E ×250).

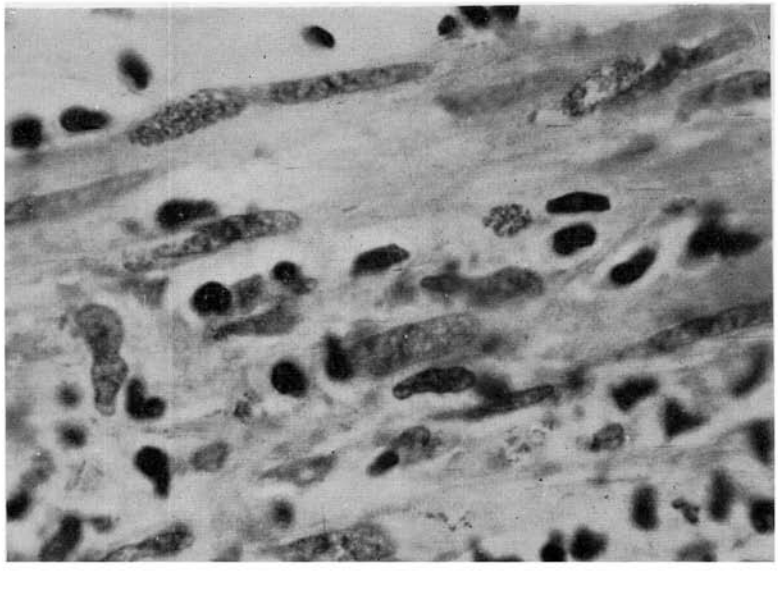

FIG. 6
Photomicrograph showing inflammatory reaction around degenerating muscle cells parasitized by *Myco. leprae* (H & E ×2000).

the muscle cells were destroyed and replaced by hyalinized fibrous tissue. Acid-fast stain showed numerous smooth muscle cells filled with bacilli, most of which were well stained and rod-shaped. Some of the bacilliferous muscle cells showed hardly any degenerative changes and elicited no inflammatory cell reaction, but in many others there were degenerative changes and granulomatous inflammation (Fig 6).

Electron-microscopic findings

The electron-microscopic studies confirmed the findings seen under the light microscope. In the striated muscle lesions the bacilli were present inside tissue histiocytes and muscle fibres. There was also focal necrosis of individual muscle fibres, which were destroyed by the presence of clusters of bacilli. Cross striations were well preserved in most of the other muscle fibres, even in those adjacent to the granuloma. There was proliferation of satellite cells. The electron-microscopic study of dartos muscle confirmed the intracellular presence of a large number of bacilli inside phagocytic vacuoles in many muscle cells (Fig. 7). Most of the bacilli were rods and were apparently viable. There were a number of mitochondria showing cystic dilatation (Fig. 8). Some muscle cells containing the organisms showed well-marked degenerative changes, with fragmentation and condensation of myofibrils (Fig. 9) and formation of myelin figures.

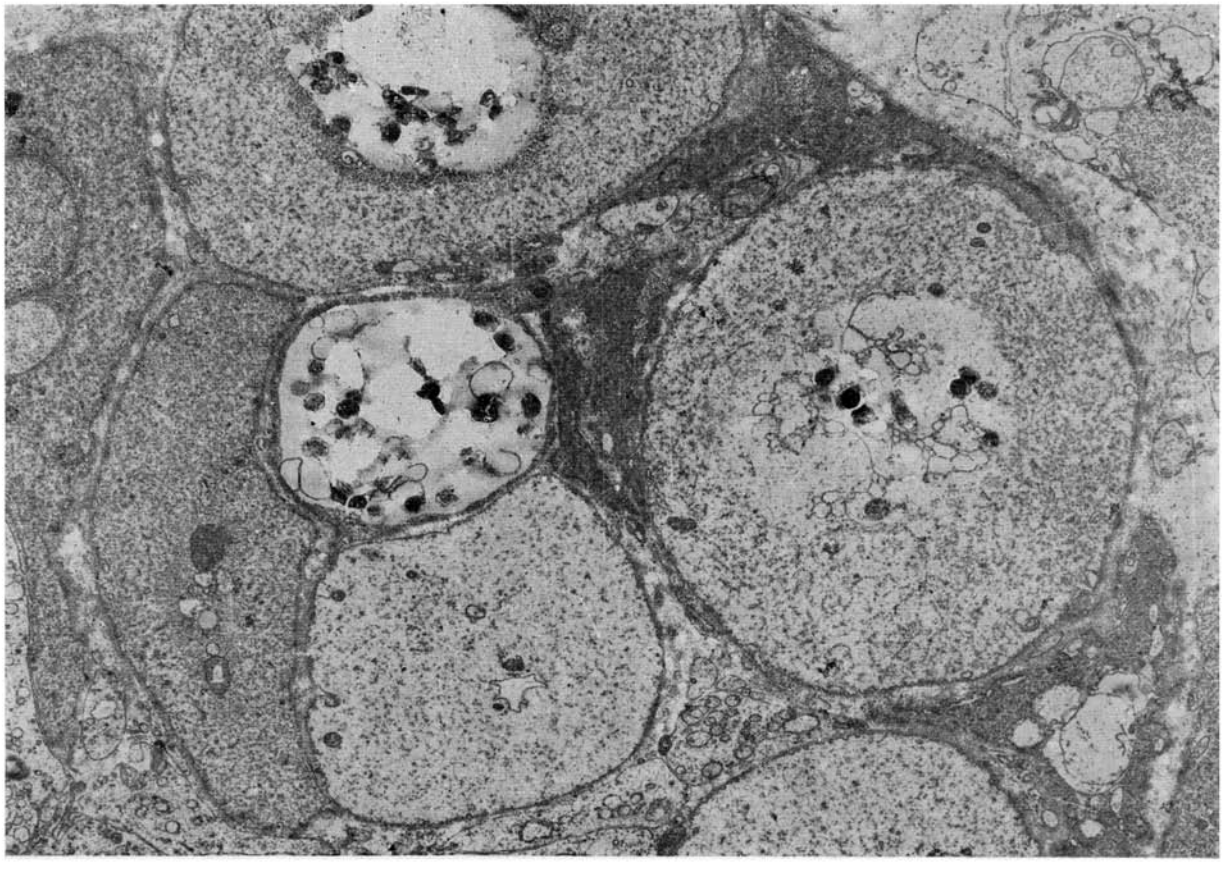

FIG. 7
Electron micrograph showing clusters of *Myco. leprae* within smooth muscle cells. Note the phagocytic vacuoles in the cells containing the organisms (× 11,900).

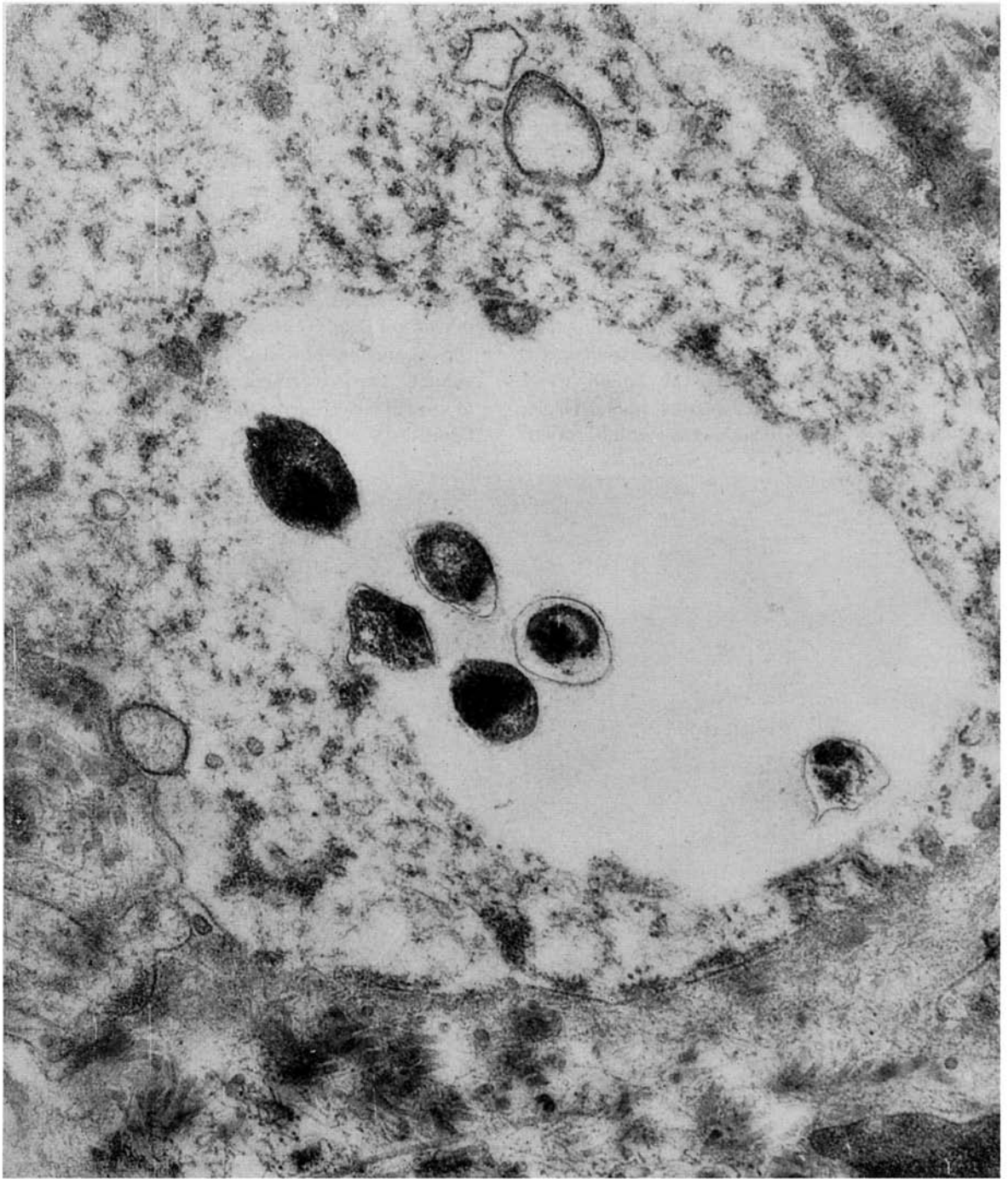

FIG. 8

High magnification electron micrograph showing several *Myco. leprae* contained in a phagocytic vacuole in a smooth-muscle cell. The mitochondria are vesiculated and the myofibrils are clumped together. Note the cross-section of the bacteria with cytoplasm, cell membrane, and the waxy coat ($\times$46,300).

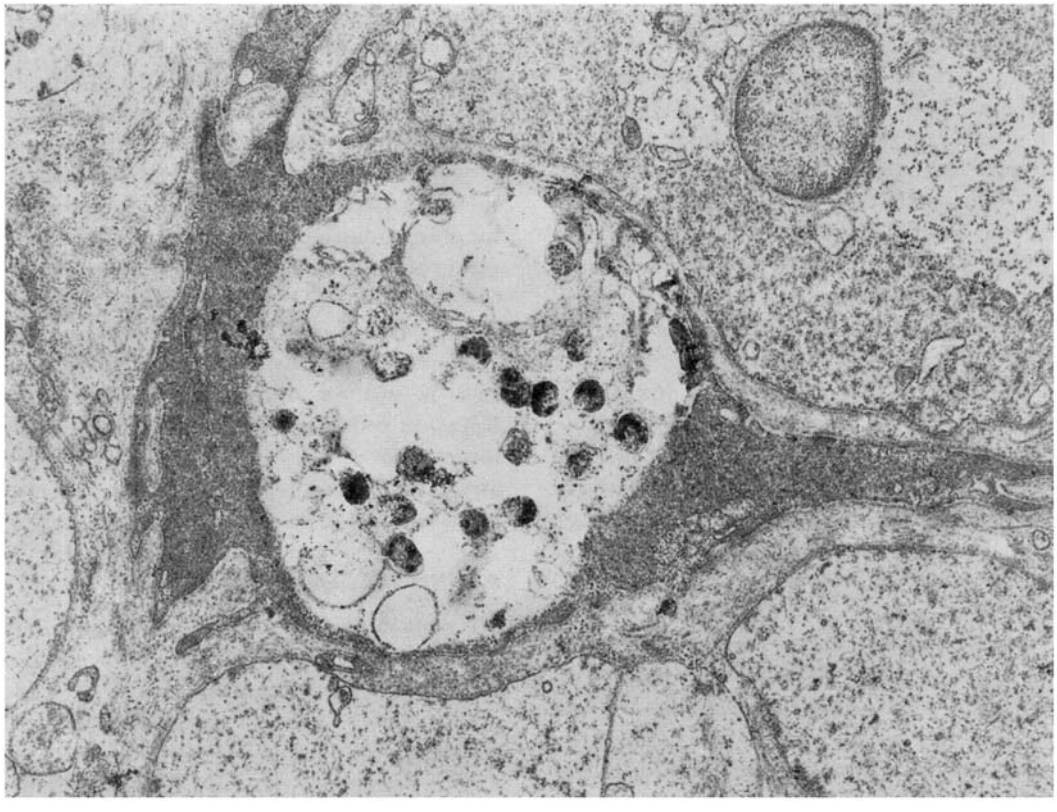

FIG. 9
Electron micrograph of a degenerating smooth-muscle cell containing a cluster of *Myco. leprae*. Note the large phagocytic vacuole almost filling the cell. The myofibrils cannot be identified any longer, but have clumped together into an amorphous electron dense granular material (× 28,900).

DISCUSSION

The common striated muscle lesion in leprosy is neural in origin, following the loss of nerve supply to the muscle. Direct invasion of the striated muscle by lepromatous granuloma has been reported to be extremely uncommon. There may be several reasons for this. Muscular lesions of bacterial origin are rare, because the muscle fibres contract so frequently that micro-organisms do not easily settle in muscle tissue and produce lesions, except in unusual circumstances. Further, the metabolites produced in muscle may alter the tissue in such a way as to prevent the growth of these organisms. Of the 6 cases in which granulomata were present in striated muscles, solid forms of *Myco. leprae* were absent in all but 2, and even in these 2 only a few solid bacilli were seen.

In the biopsy specimens from the scrotum large clusters of acid-fast rods were present inside smooth muscle cells. These latter cells degenerated into vacuolated and later necrotic cells. Smooth muscle involvement is not an uncommon finding. In skin samples from lepromatous patients it is not unusual to find the fibres of the arrectores pilorum muscle parasitized extensively by *Myco. leprae* and these organisms are also seen in the smooth muscles of blood vessels in the skin and subcutaneous tissue (C. K. Job, personal observation). Smooth muscle may have a composition which is different from that of striated muscles and

which is conducive to bacillary growth, or the smooth-muscle cells are present in an environment conducive to the growth of the bacilli. *Myco. leprae* have never been detected in the smooth muscle of the gastro-intestinal tract or other deeper structures. Therefore, it is reasonable to deduce that it is the environmental factors that make the difference. One of us (C.K.J.) believes that the lowered temperature in the subcutaneous smooth-muscle cells in the scrotum and skin is an important factor for the growth of the organisms in these sites. Smooth-muscle cells have a long life-span and their physical environment in the skin and scrotum compares well with the Schwann cells in the cutaneous peripheral nerves and therefore they also offer suitable conditions for *Myco. leprae* to multiply. We have every reason to believe that bacilli proliferate as much in the smooth-muscle cells at these sites as in Schwann cells. Therefore, it is reasonable to say that Schwann cells need not necessarily be the main target cells, as is thought at present. Any cell that has a long life-span and has an environment conducive to the growth of the organism will be parasitized and colonized by *Myco. leprae*.

The infection is carried to the muscle either through direct extension from the skin or through the blood stream. The spread along the perimysium may give the impression that the infection might have spread from the contiguous skin to the muscle along the muscle spaces. However, the special predilection of the granuloma for a site around blood vessels and the focal and selective destruction of muscle fibres following the granulomatous inflammation suggest that the bacteria are carried into the muscle bundles via the blood stream.

SUMMARY

In this study of 8 muscle biopsy specimens from lepromatous patients, of which 6 were from striated muscles and 2 from the dartos tunica of the scrotum, lepromatous granuloma consisting of macrophages containing *Mycobacterium leprae*, lymphocytes, and plasma cells were present in all of them. The bacilli were seen to grow in the muscle cells also, bringing about their degeneration and necrosis, followed usually by granulomatous inflammation. These findings were confirmed by electron-microscopic studies. Colonies of solid bacilli were demonstrated inside muscle cells. It is pointed out that in lepromatous leprosy infection of smooth-muscle cells is not an uncommon finding and that Schwann cells need not necessarily be the main target cells of *Myco. leprae*, which may grow in any cell having a long life-span and an environment conducive to the growth of the organisms.

ACKNOWLEDGEMENTS

We should like to thank The Leprosy Mission, London, and The American Leprosy Mission, New York, for making funds available for this study. Also to express our appreciation to Mr. K. George William for secretarial help, and to Mr. S. Jesudass, Mr. R. Shanthakumar and Mr. Joseph for technical help.

REFERENCES

CONVIT, J., ARNELO, J. J. and MENDOZA, S. (1960). Lepromatous myositis. *Int. J. Lepr.* **28**, 417.

HARMAN, D. J. (1968). *Mycobacterium leprae* in muscle. *Lepr. Rev.* **39**, 197.

ISHIHARA, S. (1959). A study of myositis interstitialis leprosa. *Int. J. Lepr.* **27**, 340.

PALMER, E., REES, R. J. W. and WEDDELL, A. G. M. (1965). Site of multiplication of human leprosy bacilli inoculated into the footpads of mice. *Nature, Lond.* **206**, 521.

REES, R. J. W. and WEDDELL, A. G. M. (1968). Experimental models for studying leprosy. *Ann. N.Y. Acad. Sci.* **154**, 214.

SHEPARD, C. C. (1962). Multiplication of *Mycobacterium leprae* in the footpads of the mouse. *Int. J. Lepr.* **30**, 291.