wards intraneural antigens can, however, lead to nerve damage ([10]). Since *M. leprae* or its antigens can be found intraneurally, it is possible that this mechanism is relevant in leprosy. We have sensitized a rabbit with *M. leprae* and then injected *M. leprae* sonicate into the sciatic nerve. Histologically the nerve damage seen in the injected nerve was strikingly similar to that of human nerves during reversal reaction. Whole intraneural *M. leprae* per se may not cause nerve damage, but intraneural antigens of *M. leprae* in the face of systemic delayed type hypersensitivity to them can certainly lead to a neuropathy.

In summary then, we feel that there are two completely different mechanisms involved in leprosy neuropathy. One is an autoimmune granulomatous reaction secondary to interactions between *M. leprae* and Schwann cells of unmyelinated cutaneous fibers. This reaction leads to loss of pigment and hair as well as sensory loss. The other one is a consequence of delayed type hypersensitivity to intraneural *M. leprae* antigens and affects motor or major peripheral nerve trunks. The distinction of the two mechanisms offers a more rational approach to the understanding of the immunopathology of nerve damage in leprosy and also in other diseases like diabetic neuropathy.

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Cultivation, the Neglected Priority

To the Editor:

In a recent editorial, Hastings ([2]) presented a summary of the original articles and current literature sections of the 1980 Journal. The areas of progress and frustration were clearly pointed out by the Editor. The reflections of a reader on these areas of progress and frustration prompted this correspondence.

It is instructive to express the trends in leprosy research in terms of the numbers of articles published in the 1980 Journal, tabulated according to disciplines and based on Hastings' editorial:

	Original Articles	Current Literature
Immunology	21	22
Clinical and pathology	16	17
Epidemiology	9	6
Bacteriology	7	2
Therapy and pharmacology	3	13
Mycobacterium lepraemurium	2	6
Miscellaneous	4	3
Cultivation of *M. leprae*	0	1
Total number of articles	62	70

Out of the 62 original publications cited in the editorial, none dealt with cultivation of *Mycobacterium leprae*. In the 70 articles which appeared in the Current Literature section and which were cited in the editorial, only one dealt with cultivation, and this brief article described three unsuccessful cultivation trials of *M. leprae*. Thus, both the Original Articles and the Current Literature sections of the JOURNAL reflected minimal activity in the field of cultivation of *M. leprae*.

A similar trend is evident from the abstracts of several international meetings of leprologists, such as those in Bombay (16–18 January 1981), Geneva (1–3 May 1981), Washington (13–16 July 1981), Mexico City (4–6 May 1981), and Paris (4–5 September 1981). The Paris meeting was devoted to hard-to-grow mycobacteria and the "noncultivable" *M. leprae*. In these meetings, results were presented on mass spectrometry, the ultrastructure of polar lipids, carbon metabolism, and serologic characterization of *M. leprae*, but cultivation of *M. leprae* was not discussed. In my judgment, the cultivation of *M. leprae* is the most neglected field of leprosy research. In full agreement with Hastings' analysis, the 1980 JOURNAL reflected extensive areas of progress and abundant new information. It is tempting to speculate on the impact of successful cultivation of *M. leprae* on the work presented in the 1980 JOURNAL.

If *M. leprae* were to be cultivated, for example, Kazda, *et al.* [3] could show with absolute certainty whether *M. leprae* propagate outside humans or animals in nature. Their revolutionary concept could easily be verified by simple cultivation techniques, with important implications for the epidemiology of leprosy. Seydel, *et al.* [4] could show that dapsone acts as an inhibitor of the folate synthesizing enzyme, not only in *E. coli*, but in *M. leprae*. Convit, *et al.* [1] could use heat-killed *M. leprae* from *in vitro* cultures in their vaccination procedures. This would ensure that their vaccine contained no extraneous components derived from host tissues currently used to produce *M. leprae*. Shepard, *et al.* [5] could have avoided having to perform their "meticulous and extensive studies" on the effects of purification of *M. leprae* on the immunogenicity of the bacilli for immunizing mice.

In short, the cultivation of *M. leprae* would provide an invaluable tool for specialists in leprosy bacteriology, immunology, pathology, pharmacology, epidemiology, and for the clinician. Drug-sensitive and drug-resistant cultures of *M. leprae* would serve as pharmacological models for the rapid *in vitro* screening of thousands of compounds for potential antileprosy activity. Easy cultivation might well lead to early diagnosis and the early detection of drug sensitivity or resistance, to the great benefit of leprosy patients and their contacts. Cultures of *M. leprae* could provide unlimited quantities of bacilli for antigenic analysis, for the preparation of specific vaccines, for lepromin skin tests, and possibly for an antigen for serodiagnostic purposes. On a more practical level, one would expect that cultures of *M. leprae* could provide for the production of bacteria much more economically than they now have to be produced by the time-consuming and expensive procedures employing armadillos.

Clearly then, the cultivation of *M. leprae* is a highly desirable goal and one which would be expected to yield large benefits. Why, then, is cultivation not given more priority in leprosy research? The number of "cultivators" is diminishing year by year. Only a few laboratories in the world are now engaged principally in cultivation trials of *M. leprae*. Is cultivation of *M. leprae* really a "mission impossible"? Are we accepting too readily the traditional concepts of *M. leprae* as "obligate intracellular par-

asites" or "noncultivable mycobacteria" or "metabolically deficient" organisms? Certainly a great many highly qualified scientists have devoted years of their lives to pursuing this elusive goal without success. Do younger researchers feel that their careers will be threatened if they do not produce fast results? Are the controversies which have surrounded previous claims of cultivation of *M. leprae* discouraging younger investigators from taking up the task? Responsible persons and agencies must find a way to encourage research on the cultivation of *M. leprae*. What an exciting and rewarding field this is, to force that elusive microorganism to split in a test tube! Millions would benefit in a not-too-distant future from the discovery. Are we guilty of scientific negligence if we fail to continue to emphasize this goal?

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Isolation of Fastidiously Growing Mycobacteria from Armadillo Livers Infected with *Mycobacterium leprae*

TO THE EDITOR:

In 1980 we received four armadillo livers infected with *Mycobacterium leprae*. Their origin is indicated in Table 1. The specimens were transported on dry ice and kept in the laboratory in an electric deep freeze at −70°C until use.

Specimen I was from an animal with a "natural" *M. leprae* infection ([18]), while the other three were from animals injected with *M. leprae* extracted from human biopsy material.

In our laboratory each liver was aseptically divided into different samples. Suspensions in distilled water (50% w/v) were prepared and investigated on different dates (Table 1). The viability of the acid-fast bacilli (AFB) was tested by titrations in mouse foot pads (MFP), starting with an inoculum of 5×10^3 per foot pad. Animals were examined tri-monthly from the sixth month on, and five of them killed and counts performed when 5×10^5 AFB/MFP was reached; the last examinations were performed one year after inoculation.

Two-tenths ml amounts of the same suspensions were inoculated on the surface of slants of modified Ogawa medium ([10]) either without prior treatment or after addition of different concentrations of hydrochloric acid or sodium hydroxide (final concentrations from 0.1% to 2% and neutralized or not). Modified Ogawa medium was prepared by the addition of autoclaved suspensions of one of the following mycobacterial species: *M. phlei*, *M. lufu* ([12]), *M. lepraemurium* ([10]) or *M. leprae* (from human biopsies or armadillo livers) and adjusted to different pH values ([15]). Incubation was at 30°, 33°, and 37°C, with or without CO_2 and in incubators, humidified or not, stoppers pierced with 19G needles or not ([10, 14]), for a period of one year.

Results of titrations in MFP revealed that