pads was noted. In the foot pads of nude mice infected with $1 \times 10^5$, $1 \times 10^6$ and $1 \times 10^7$ bacilli, maximum yields of $1.7 \times 10^{10}$, $2.0 \times 10^{10}$ and $2.1 \times 10^{10}$ bacilli per foot pad were estimated, respectively, after 13, 12 and 11 months of infection. Dissemination of *M. leprae* in foot pads of nude mice is well established (7-3). These results of a comparative study show that, like nude mice, SCID mice were also susceptible to *M. leprae* infection and the onset of the lepromatoid lesions in the foot pads of SCID mice occurred earlier than that observed in the nude mice. However, an interesting aspect of this study is that the progress of *M. leprae* infection in SCID mice is different than the progress observed in the nude mice. Although a rapid multiplication of *M. leprae* occurred in the foot pads of SCID mice after reaching a maximum of about 4 to 9 $\times 10^8$ bacilli/foot pad, the number of bacilli decreased upon further incubation and eventually, about 15 months postinfection, only a few degenerated bacilli were found at the site of infection and there was no sign of dissemination. On the other hand, the number of bacilli in the foot pads of nude mice after reaching about 1.5 to 2.0 $\times 10^{10}$/foot pad remained nearly the same up to 15 months of incubation. Since the total number of bacilli in the foot pads of nude mice is considerably higher, such mice should continue to be used for the production of *M. leprae* for leprosy research. SCID mice possibly could be used for screening the antileprosy drugs in a relatively shorter period of time.

The infection of *M. leprae* in SCID mice has not been investigated extensively. The phenomenon of decline and eventual clearing of millions of bacilli in the foot pads of SCID mice in a relatively shorter time period is not clear and is worthy of investigation. Additional studies such as histopathology and the status of the immune response of SCID mice at various stages of infection, lepromin reactivity as well as DNA homology, should be carried out.

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REFERENCES

Accurate Diagnosis of Tuberculosis Meningitis Using Polymerase Chain Reaction

TO THE EDITOR:

Tuberculous meningitis (TBM) is unique and important in the pediatric age group and happens to be the most common cause of death in children suffering from tuberculosis. Favorable prognosis depends upon the early diagnosis of tuberculous meningitis, for which reliable methods based on serology are not available. The ultimate diagnosis for TBM depends on isolation and identification of mycobacterial species, which is time-consuming and often gives negative results in spite of clinical disease. We previously have reported the presence of a repetitive sequence on 5.6-Kb AluI restricted *Mycobacterium tuberculosis* DNA
THE TABLE. Effectiveness of polymerase chain reaction in detecting early stages of TBM.

<table>
<thead>
<tr>
<th>No.</th>
<th>AFB+</th>
<th>Culture+</th>
<th>PCR+</th>
</tr>
</thead>
</table>
| Group I—Controls
| Category of patients | | | |
| Pyrogenic meningitis | 8 | 0 | 0 | 0 |
| Aseptic meningitis | 4 | 0 | 0 | 0 |
| Enteric encephalopathy | 4 | 0 | 0 | 1 |
| Other neurological disorders | 9 | 0 | 0 | 0 |
| Total | 25 | 0 | 0 | 1 |
| Group II—TBM Cases
| Clinical stage | | | |
| I | 2 | 0 | 0 | 2 |
| II | 6 | 0 | 0 | 5 |
| III | 17 | 1 | 3 | 15 |
| Total | 25 | 1 | 3 | 22 |

(4). Subsequently, a polymerase chain reaction (PCR)-based test which amplifies the 169-bp DNA fragment unique to the *M. tuberculosis* complex from a single-copy region lying adjacent to the repeat element of 5.6-Kb Alu fragment recently has been reported by us (4). This report describes the evaluation of a PCR test on suspected cases of tuberculous meningitis in a blind manner.

Fifty patients, including 25 controls, attending the pediatric ward of Lok Nayak Jai Prakash Narayan hospital, New Delhi, took part in the study. Twenty-five patients were suspected for TBM on the basis of criteria such as fever, altered sensorium, headache, persistent vomiting, convulsions, neurological deficit, nuchal rigidity and positive history of contact with tuberculosis. CSF analysis showed predominant lymphocyte pleocytosis with protein content over 50 mg% and sugar less than 45 mg%. The controls included 25 patients having pyogenic meningitis, aseptic meningitis, enteric encephalopathy and other neurological disorders. Samples were processed in duplicate, and PCR amplification was carried out with primers KS3-5'CAACCAACATCGCTGAC; KS4-3'ATAGCGGATGACGGTCG 5' according to Reddi, et al. (4). At the end of the study the results were compared with the clinical histories of the patients along with culture and smear results. Figure 1 represents an autoradiograph of some of the samples. Figure 2 represents a typical CAT scan finding of a suspected TBM patient. The Table details cumulative results obtained on controls and patients.

Of 25 suspected TBM cases in different stages, a 169-bp amplified product specific to *M. tuberculosis* was detected in 22; only 1 was positive by the smear test and 3 were positive by culture tests. Those patients found positive by the PCR test had corroborations with a clinical picture as well as a CAT scan. PCR negativity in three confirmed TBM patients could be due to a low bacillary load with the clinical manifestation being due to a hypersensitivity reaction. The effectiveness of the PCR test in finding infection in the early stages of the disease is evident (The Table) although studies were carried out on a limited number of samples due to their unavailability. Out of 25 control specimens only one
showed false positivity, which could be due to laboratory error. During the past couple of years, several reports have appeared describing the PCR tests (1-5) and their evaluation of specimens from pulmonary and extrapulmonary tuberculosis cases. Subsequent reports regarding misdiagnosis using the PCR/DNA probe test have appeared (6). The importance of the PCR test in diagnosis is undisputed, and erroneous results occasionally obtained could be due to strain variation-laboratory errors. Thus, it appears important to use more than one set of primer combinations while confirming the diagnosis. The PCR test developed by us initially was evaluated in a blind trial on limited sputum specimens and was found to be 100% accurate with a concordance rate of 76%. The importance of this test is emphasized by the data presented in this communication.

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Mycobacterium lepraemurium Itself Does Not Interfere with the in Vitro Lymphoproliferation Induced by Concanavalin A

TO THE EDITOR:
Mycobacteria are microorganisms that penetrate the macrophages by active phagocytosis, and phagocytic killing is the only mechanism known for the “physiological” destruction of these bacteria (6). Macrophages, however, although efficient in general terms, have a limited capacity to destroy pathogenic mycobacteria such as Mycobacterium leprae and M. tuberculosis. The microbicidal activity of macrophages is highly potentiated by the effects of cell-mediated immunity (CMI) through the several lymphokines produced, among which gamma-interferon (IFN-γ) is of particular relevance. IFN-γ-activated macrophages exert clear deleterious effects on the leprosy bacillus (7).

Patients with lepromatous leprosy develop a specific loss of CMI to M. leprae antigens, and although the reason for this is still not well understood several tentative explanations have been put forward. Following infection, M. leprae cells are cap-