The Present State of Growth of *M. leprae* Under *in Vitro* Conditions

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For the *in vitro* cultivation of *M. leprae* special double tubes, (U-shaped tubes) separated by a sintered glass filter plate (Schott G 5) at the middle, were used. Each side of the tubes contained the growth medium; one side served for the inoculation and the other for the current replacement of the medium, thereby, in contrast to normal test tubes, functioning in a simple way as an open system (Fig 1).

A great number of specimens from untreated patients with lepromatous leprosy containing masses of acid-fast organisms were investigated for *in vitro* growth experiments. Several techniques, as, for instance, separation of leprosy bacilli from tissue specimens, pretreatment of the specimens with sulfuric acid or other agents, preparation, composition, and replacement of the media used, inoculation of leprosy bacilli as well as their staining and counting after different times of incubation, will be described and discussed in detail.

After an incubation time of nine to 12 months limited multiplication of *M. leprae* was observed in 15.8 per cent of the cultures. The range of temperature in which growth occurred was very small. The optimal temperature for growth was 32° C to 33° C. The generation time (G), calculated according to the equation

\[ G = \frac{1}{\log N_t - \log N_0} \]

varied between 20.8 days and 41.3 days with an average of 28 days. Of the cultures 84.2 per cent showed no multiplication of *M. leprae* because of contaminations, blocking of the filter pores, or other reasons.

† Deceased.