# REPRINTED ARTICLE

# Some Recent Laboratory Findings on Mycobacterium leprae

## Implications for the Therapy, Epidemiology and Control of Leprosy

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A recent guide to a national leprosy control programme stated that even infectious cases of leprosy can be rendered non-infectious in a period of 4-6 months, that the morphological index (MI), i.e., the percentage of solid-staining bacilli present, falls to practically 0 in 4-6 months although smears usually remain positive, and that, from the public health point of view, treated leprosy patients are no longer a source of infection to the community. At the East African Leprosy Conference meeting at Kampala, Uganda, in April 1970 (unpublished documents) two of the resolutions unanimously adopted were as follows:

"It has been known for many years that only a minority of leprosy cases are infectious, and it is now also known that the infectivity of these cases is rapidly reduced to nil by treatment . . ."

"Most leprosy patients are not infectious and recent studies have shown that the majority of smear-positive patients become non-infectious within three to four months of the beginning of regular treatment."

These views are based on the findings of a series of laboratory studies on the morphology and viability of Mycobacterium leprae in mice and, if they were widely adopted, public health administrators and leprologists would be induced to devote less time to epidemiological surveillance, to cut down on control measures and to reduce the strength of leprosy programmes. The consequences could be serious for the populations of endemic leprosy areas and the views accordingly require careful examination.

### LABORATORY FINDINGS

The importance, and far-reaching consequences, of recent laboratory findings in leprosy may be illustrated by some quotations from the recent literature. Rees & Waters (1963) stated:

"The most recent studies . . . clearly demonstrate for the first time that irregularly stained forms seen in human leprosy bacilli can be identified exactly with degenerate forms of bacilli seen with the electron microscope, and from previous experimental studies with murine leprosy bacilli and with E. coli, these irregularly stained organisms are likely to be nonviable. It is reasonable, therefore, to conclude that only the uniformly staining human leprosy bacilli, the so-called 'solid' forms, are likely to be viable. All forms of irregularly stained human leprosy bacilli, whether defined as 'fragmented' or 'granular' or 'beaded' can be considered dead organisms."

Based on their studies, the authors believed that more than 50% of the leprosy bacilli in untreated patients were already dead and stated:

"In human leprosy as in murine leprosy, bacilli can be readily killed but only very slowly disposed of by the tissues."

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In another paper, Rees (1969a) has stated:

"It has long been considered that even the most active antileprosy drugs, including diaminodiphenylsulfone, leave much to be desired since in patients with the more severe lepromatous type of leprosy many years elapse before negative smears give evidence of a 'cure.' Hitherto, it has been assumed that more active drugs are required that would kill Myco. leprae more rapidly than diaminodiphenylsulfone, an assumption that was based on the fact that the BI [bacteriological index] falls so slowly. Now that it has been demonstrated conclusively that the viability of Myco. *leprae* can be assessed on the basis of their morphology rather than on the total number of the bacilli, it has been possible to show, in carefully controlled studies, that a very high proportion of bacilli are killed in 3 months in patients receiving standard treatment with diaminodiphenylsulfone (Waters & Rees, 1962). This observation suggests that persisting lesions and many of the manifestations of leprosy, including reactions of the erythema nodosum leprosum type, that follow the initial phase of chemotherapy must be due, in part, to the presence of dead bacilli. It implies that a more rapid cure will be achieved only if other drugs or methods are found that could be used, after the inital killing of leprosy bacilli with standard antileprosy drugs, to enhance the host's ability to dispose of dead, but still intact, leprosy bacilli."

In the same paper Rees (1969a) also stated:

"It is clear . . . that a new and valuable technic was available that could now be used in man to determine the viability of *Myco. leprae* in patients and thus to assess precisely their probable response to chemotherapy, the infectivity of the various types of leprosy, and, by histological studies, the viability of *Myco. leprae* in different tissues. Thus the morphological index (MI), that is, the percentage of solidly staining bacilli seen in smears or sections from leprosy patients, could now be used as the measure of viability."

The limited multiplication of Myco. le-

prae in the footpads of mice was first reported by Shepard (1960) and he subsequently (Shepard, 1969) observed that the footpad "infection" was directly related to the number of solidly staining Myco. leprae in the inoculum rather than to the total number of bacilli. About 5000 bacilli were usually inoculated into a footpad and this number included as many as 400 viable organisms, judging by the number that were "solid." The bacilli multiplied only up to a plateau level of about  $2 \times 10^6$  in 6 months (Shepard & McRae, 1965); tissue changes in the footpads were minimal and microscopic enumeration was required to detect actual multiplication.

Shepard, Levy & Fasal (1968) reported that 3 months of treatment with diaminodiphenylsulfone<sup>1</sup> was sufficient to kill more than 99% of the "viable" bacilli in lepromatous patients.

"The infectiousness for mice decreased in the first 30 days, and was only barely detectable at 30 to 90 days, after which it was not detectable. In the range where it could be determined accurately, the proportions of solidly staining bacilli decreased in parallel with the infectiousness. The number of bacilli in skin specimens [obtained by repeated biopsies] decreased much more slowly during the period of observation (the first 300 to 400 days of treatment). The rate of loss was estimated to average 0.93  $\log_{10}$  units (8.5-fold) per year."

"The measurements of infectiousness were sensitive enough to follow the loss of viability down to a level at which less than 1% of the starting value remained. However, to effect a bacterial cure (so that patients will not relapse when treatment is stopped) it is presumably necessary to kill all the viable organisms. Most lepromatous patients probably have a total of about  $10^{11}$  leprosy bacilli, or  $10^9$  to  $10^{10}$  viable bacilli. Obviously the present results apply only to the first stages of antibacterial therapy and do not allow one to predict the duration of treatment required for a cure. However, the present results do emphasize

<sup>&</sup>lt;sup>1</sup> Proposed international non-proprietary name: dapsone.

how greatly reduced are the chances for the lepromatous patient to infect others after even a few months of treatment. Presumably the infectiousness in these patients would be reduced in this period to less than 1% of the pre-treatment level."

These conclusions were drawn from a laboratory study of the changes in morphology and resulting viability in mice of bacilli taken by skin biopsies and nasal washings from only 6 lepromatous patients under treatment with dapsone for 300-400 days. Two of the patients, moreover, had already received dapsone therapy—namely, 300 mg/day for 71 days and 100 mg/day for 31 days. At the start of the study, the MI from skin biopsies of the 4 untreated patients were: 9%, 4%, 2% and 2%. "Usually 50 bacilli were scored when the MI was estimated" (Shepard, Levy & Fasal, 1968).

With respect to the minimal effective dose of dapsone, Shepard (1969) stated that, "Recent evidence stemming from investigation of the minimal effective dosage against M. leprae in mice indicates that even the standard dosage (50 to 100 mg per day) provides 50 to 100 times the minimal inhibitory concentration." Shepard was referring to the paper by Waters, Rees & Ellard (1968) in which it was stated that "... wild strains of M. leprae from different parts of the world were exquisitely sensitive to DDS [dapsone], being inhibited by a concentration in the diet that gave serum levels of 0.01-0.02  $\mu$ g/DDS/ml. It was estimated that such levels would correspond in man to a daily dose of 1 mg DDS". This finding in mice was presumably confirmed by an actual trial in 7 previously untreated lepromatous patients who were given 1 mg of dapsone daily for 4-5 months. The sensitivity to dapsone of the bacilli in each patient was determined by successive inoculations in mice. Interim clinical and bacteriological results were "encouraging" but Waters, Rees & Ellard (1968) did not advocate the general use of the 1-mg dose of dapsone because of the risk of drugresistant strains of Myco. leprae developing.

The effectiveness of BCG against Myco. leprae in mice was reported by Shepard (1968) as follows:

"Following inoculation into the footpad of a normal mouse, M. leprae multiplies with great regularity. . . . The bacillary growth curve consists of a lag phase, a logarithmic phase, and a plateau phase, and advantage can be taken of the regularity of the curve to measure relatively small effects. The end of the logarithmic phase, and thus the onset of the plateau phase, is apparently a function of the immune response in the normal mouse to a bacterial population of about 106 bacilli and at this time many M. leprae are killed. In a mouse that has been well vaccinated with BCG, or with oil-treated BCG cell walls, the immune response is triggered by a lower bacterial population and the plateau level is 10- to 100-fold lower."

In another paper (Shepard & Ribi, 1968) it was stated:

"BCG, an anti-tuberculosis vaccine, provides protection against the footpad infection in mice with M. leprae. Although ordinary preparations of BCG cell walls are not effective as vaccine against tubercle bacilli, oil-treated cell walls provide good protection against pulmonary infections of mice with M. tuberculosis. Similarly, we have found that oil-treated BCG cell walls were effective as vaccines against M. leprae infections in mice. The amount of protection was about as great as that provided by intact (living) BCG, and was distinct, following either intradermal or intravenous administration. Cell walls that had not been treated with oil provided no protection."

Referring to Shepard's BCG experiments in mice, Rees (1969b) concluded that the ". . . experimental infection has provided indisputable evidence of the immunizing capacity of BCG vaccine against the leprosy bacillus".

Thus, laboratory findings in mice have been directly applied to human leprosy and if, in fact, they are applicable, they would entail a complete change in the methods of treatment and prevention. An effect in one animal species may not, however, apply to man. *Myco. leprae* "infections" in the footpads of mice, limited at best, die off after reaching a certain level, indicating that mice and humans are not alike in their susceptibility; man (mainly the non-reactor to lepromin) is naturally susceptible to *Myco. leprae* and can develop lepromatous leprosy; mice, unnatural hosts, may not react in the same way.

Dapsone has been used very extensively in the treatment of leprosy for about 20 years; BCG vaccination has been used very extensively in tuberculosis control programmes, but less widely in connection with leprosy. Judged by their great effectiveness in leprosy in mice, these two measures together should by now have accomplished a world-wide reduction in the prevalence of leprosy but there is no evidence that such a reduction has taken place.

Subsequent events may confirm the importance of the laboratory findings on *Myco. leprae* in mice but it would be premature to apply them to human leprosy until definite confirmation is obtained from careful clinical and epidemiological studies in man.

## LABORATORY FINDINGS AND LEPROSY THERAPY

It has already been stated that Myco. leprae in mice were found to be so sensitive to dapsone that the equivalent minimal effective dose in man would be as low as 1 mg daily. As a result, the use of very low dosages of dapsone has actually been recommended in some current leprosy control programmes. However, none of the field trials so far reported in support of low dosages is fully acceptable. Among their other limitations, these studies were not double-blind trials or did not make use of controls.

Karat, Jeeveratnam & Rao (1969a) reported an admittedly limited study in 20 lepromatous cases treated for at least 1 year with 5 mg or 10 mg of dapsone daily. These authors noted that the incidence of erythema nodosum leprosum seemed to be little affected but that the severity of the reactions was generally less. There was little or no change in the morphological index but the bacillary index showed a disturbing tendency to rise. None of the 20 patients showed any significant improvement in the appearance of the skin lesions. They also commented:

"The long-term results of this study would indeed be disturbing if they suggested the appearance of resistant strains of bacilli. One fervently hopes that this will not happen because it would be a major disaster in the natural history of leprosy as a whole. In countries where there is no other drug available than dapsone for the general management of leprosy, appearance of resistance to this 'sheet-anchor' of treatment would be an unmitigated disaster! Therefore it would be in order for us to sound a word of caution against the general recommendation of the use of very small doses of dapsone in the routine management of leprosy. If one could restrain the therapeutic enthusiasm dictated by changing fashions, one might obtain more satisfactory longterm results!"

Further details on this study were given by Karat, Jeeveratnam & Rao (1969b). They inoculated bacilli from biopsies from patients on low doses of dapsone into the footpads of mice at periodic intervals and stated that ". . . data available to date suggest that the bacilli in the skin of patients treated with small doses of dapsone are viable even after 18 months of continuous treatment." They also stressed that the presence of dapsone in the sera of these patients was confirmed by Dr. C. C. Shepard and there is thus no doubt that the patients had been given regular doses of dapsone. These authors have also indicated that further analysis of data on these patients is in agreement with the findings which they have already presented.

It may also be added that Levy (1967), referring to the paper by Shepard, McRae & Habas (1966) on the minimal effective dose of dapsone commented: "It is unquestionably extremely hazardous to extend the results in the mouse to man". Levy added, however, that their investigation ". . . certainly suggests that doses of dapsone in man a good deal smaller than the traditional 300-600 mgm/week may well be therapeutic . . . ". In fact, sufficient evidence may be cited to show that lower than standard doses of dapsone may be useful although less effective in bringing about clinical and bacteriological improvements.

Quagliato, Bechelli & Marques (1968)

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Clinical cate- gory of lepromatous cases	Regular treatment								Irregular treatment						
	No.	Cumulative coefficients at the end of the following numbers of years:							Cumulative coefficients at the end of the folloing numbers of years:						
		1	2	3	5	9.5	≥10	No.	1	2	3	5	9.5	≥10	
L1	193	34	62	79	91 74	96	97	30	20	43	60	69	84		
L2 L3	286 211	4 0	23	46	44	94 83	99 97	49 46	0	0	4	15	80 59	92 74	

TABLE 1. Cumulative coefficients of bacterial negativity in lepromatous patients treated regularly with dapsone and in those treated irregularly;<sup>a</sup> Campinas, Brazil, 1948–67.

<sup>a</sup> Irrespective of any subsequent reactivation that may have occurred.

reported an important 20-year retrospective study in Brazil concerning the acquisition of bacteriological negativity and the subsequent occurrence of relapses in 815 lepromatous patients, in relation to the regularity or irregularity of dapsone therapy. Since Brazilian regulations require a 2+ Mitsuda reaction before lepromatous patients are released from control, regardless of clinical or bacteriological status, these patients were continued under prolonged dapsone treatment even after they had become bacteriologically negative. The incidence of bacteriological negativity and of subsequent episodes of relapse were estimated by a modified life-table method and expressed, rather unusually, in terms of cumulative coefficients, as indicated in Tables 1 and 2.

Table 1 is worth noting mainly because it confirms the very common experience that 5 and even more years of dapsone treatment are required to make many lepromatous patients bacteriologically negative. This table also shows that the duration of treatment needed to achieve bacteriological negativity is influenced by the clinical state of the patient at the outset and by the regularity, or irregulatity, of dapsone therapy—in other words, the dosage. It should be appreciated that even the most irregularly treated of these patients would be considered as grossly over-treated on the basis of the laboratory findings in mice.

Because of faults common to long retrospective studies, the rates of relapse in Table 2 may be unnecessarily high. They are notable none the less, since they indicate

TABLE 2. Cumulative coefficients of reactivation (relapse) in lepromatous patients receiving dapsone treatment regularly or irregularly after becoming bacteriologically negative: Campinas, Brazil, 1948–67.

Clinical cate- gory of lepromatous cases	Regular treatment								Irregular treatment						
	No.	Cumulative coefficients at the end of the following numbers of years:							Cumulative coefficients at the end of the following numbers of years:						
		1	2	3	5	9.5	≥10	No.	1	2	3	5	9.5	≥10	
L1	163	1	1	4	8	19	27	22	0	0	5	11	34	56	
L2	235	0	1	4	12	27	29	39	0	0	6	29	56	86	
L3	183	2	3	6	14	27	37	32	0	3	7	28	62	86	

that a substantial number of lepromatous patients who become bacteriologically negative after standard dapsone therapy still relapse, especially after 5 or more years, even if dapsone treatment is continued. Table 2 also shows that the occurrence of relapses was influenced by the regularity or irregularity of dapsone treatment (i.e., dosage) which the patients continued to receive after becoming bacteriologically negative.

Quagliato, Bechelli & Marques have also reported that among 206 cases inactive ("arrested") for 2 or more years and treated for at least 5 years, 35 still had bacterial positivity and skin sections showed that 36 had lepromatous structure in regression. Altogether, 20 out of 159 patients with chronic inflammatory infiltrate and bacterial negativity in skin sections had later bacterial reactivation in skin lesions, although still under treatment with sulfones.

Studies of relapse in lepromatous patients after standard dapsone therapy have understandable shortcomings, in view of the great difficulty of following up these patients for long periods after discharge from clinics and leprosaria. Erickson (1950), Lowe (1954), Rodriguez (1958), Quagliato, Berquo & Leser (1961), Hazizov (1964) and Torsuev et al. (1965) have reported on the rates of relapse following dapsone therapy; they are unanimous in stating that relapses occur in patients rendered bacteriologically negative by prolonged, conventional dapsone therapy, although in varying, and often small, proportions (probably as a result of the limited periods of observation). In consequence, some prominent leprologists recommend life-long dapsone treatment for all lepromatous patients, or at least treatment until they become Mitsuda-positive.

Contrary to the results of dapsone therapy in mice, the shortcomings of sulfone therapy are again evident in the Chingleput chemoprophylaxis trial in India (Dharmendra et al., 1965, 1967) in child contacts, all of whom were without signs of *leprosy* at the outset. By the end of 5 years of chemoprophylaxis, leprosy had occurred in 48 out of 360 (13.3%) children in the control group, but also in no less than 23 out of 358 (7.4%) children in the dapsone group. The "prophylactic" doses of dapsone, for example, 150 mg per week for children 10-14 years of age, were more than *adequate* and very much greater than the presumed minimal effective dose, which is only 1 mg daily; these dosages did not, however, prevent apparently normal child contacts from developing leprosy.

Rees (1969) and other authors have now reported on many drugs other than dapsone, all of which have been found to be effective against Myco. leprae in the footpads of mice; they include: long-acting sulfonamides, thiacetazone, clofazimine, streptomycin, isoniazid, p-aminosalicylic acid, capreomycin; and also rifampicin, Oxydiazolone, Cephazolone, cefaloridine, rifamycin, streptovarycin and viomycin. Thus it would appear that a large number of potentially effective drugs are readily available for trials against leprosy, but a number of these drugs (including isoniazid, p-aminosalicyclic acid and long-acting sulfonamides) have already been sufficiently tested in lepromatous patients without conspicuous success. For all its shortcomings, dapsone continues to be the drug of choice in human leprosy. The viability of Myco. leprae in the footpads of mice is undoubtedly valuable for screening antileprosy drugs but the results of trials should not be applied to human leprosy without final proof in lepromatous patients.

Reactional tuberculoid cases are usually bacteriologically positive and occasionally even strongly positive; such cases may become bacteriologically negative in less than 6-12 months, even without sulfone treatment; the bacilli disappear as soon as the reactional phase subsides. Patients with borderline leprosy are almost as strongly positive as those with purely lepromatous lesions. Regardless of dapsone therapy, however, the rate of fall in a bacteriological index (BI) is 3 or more times faster in borderline than in lepromatous leprosy. Dapsone has been found to be extremely active against Myco. leprae in mice and, on the basis of the change in MI of patients under therapy, it now appears that a more rapid cure can be achieved only with the

discovery of a supplementary drug, to be used in conjunction with dapsone, that would enhance the elimination of dead bacilli responsible for the long-persisting lesions of lepromatous patients. It seems, however, that the elimination of bacilli from the body is not related to therapy but rather to the immune mechanisms of the patient, particularly the so-called "cellular" or "cell-mediated" defense mechanism. In all the forms of leprosy, lepromatous, borderline or tuberculoid, clinical improvement coincides with a fall in the BI rather than the MI, with or without therapy.

As previously stated, the MI is now being regarded by some authors as a definite measure of response to therapy, presumably without any consideration of the condition of the patient under treatment. The opinion of a group of experts who took part in a consultation arranged by WHO in London in 1968 (Convit et al., 1970) was that the significance of the MI in relation to the BI and the clinical status, as well as to histopathological findings, should be appraised in long-term clinical studies.

Viability of Myco. leprae in mice and vaccination with BCG

The potential value of BCG vaccination against leprosy has long been recognized, together with the many difficulties inherent in anti-leprosy trials with BCG. The 10-100-fold reduction in *Myco. leprae* "infection" in the footpads of vaccinated mice reported by Shepard (1968) which is now considered by Rees (1969b) ". . . some indisputable evidence of the immunizing capacity of BCG vaccine against the leprosy bacillus".

Major trials of BCG in leprosy are now in their final stages; the trials are in Uganda (Brown & Stone, 1966; Brown, Stone & Sutherland, 1968a, 1968b, 1969) and Burma (Bechelli et al., 1970). Because of their implication for leprosy prevention, the results of these large-scale, controlled studies should be analysed together. The main findings of the Uganda study are summarized in Table 3 which seems to show that BCG very substantially prevented leprosy infections: by 80% in about 2 years, 98% in 4 years and 74% in 6 years, i.e., an average reduction of 84%. Other unexpected results are not shown in Table 3. Judged only by lepromin-inducing properties, BCG its might be expected to prevent lepromatous, but not tuberculoid, leprosy. In the Uganda study, however, BCG effectively prevented tuberculoid leprosy; virtually all cases among unvaccinated children were early tuberculoid leprosy. BCG presumably conferred equal degrees of protection on

TABLE 3. Incidence of leprosy in vaccinated and unvaccinated contacts or relatives of predominantly tuberculoid cases: negative or weak tuberculin reactors aged 0-16 years, Uganda, 1962-68<sup>a</sup>.

	Fo			
	First—(up to May 1964)	Second—(up to March 1966)	Third—in prog- ress (up to June 1968)	Totals
No. of children in the trial:	8071	9036	0026	0026
BCG	8091	9052	9052	9052
No. of leprosy cases (and rates):		it - Tabah		
Control	89 (11.0%)	54(6.0%)	31 (3.4%)	174 (19.3%)
BCG	18(2.2%)	1(0.1%)	8 (0.9%)	27(3.0%)
Reduction in incidence attributable to BCG	17 St. 55			
vaccination (%)	80	93	74	84
		1		

\* From Brown & Stone (1966) and Brown, Stone & Sutherland (1968a, 1968b).

Uganda children at *all* ages, whether aged 0-9 or 10-15 years, within the 2 years following vaccination; this implies that the vaccine could have aborted pre-existing or incubating leprosy infections among the older children. BCG "infection" furthermore appeared to confer on the children a much greater protection than that attributable to actual human tuberculosis infection.

In spite of these impressive results, however, Brown, Stone & Sutherland (1968a) stated:

"The current results indicate that BCG gives about 80% protection against the early forms of leprosy in this part of Africa; it is too early to assess protection against lepromatous leprosy."

"These early results are very encouraging but personal experience in Africa since 1930 suggests that the pattern of leprosy is changing, perhaps as a result of chemotherapy. It would therefore be unwise to generalize from these results or to reach a premature conclusion."

In the WHO-sponsored Burma trial (Bechelli et al., 1968, 1970) the objective was to determine the protection conferred by BCG on the general child population (0-14 years) of a highly endemic area with an appreciable proportion of lepromatous leprosy. The trial started in August 1964; by July 1968, 12,995 children had been included in the BCG group and 12,983 matching children in the unvaccinated control group. The preliminary results up to July 1968, after 3 annual re-examinations following vaccination, can be summarized very simply: 113 cases of leprosy were detected in the BCG group compared with 128 in the control group. In both groups only early, and mainly tuberculoid, forms of leprosy were detected. So far, BCG has failed to confer any significant protection against leprosy under the conditions prevailing in Burma at the time of the trial. Perhaps, when follow-ups have been continued for another 5 years or more, the effect of BCG vaccination on the incidence and evolution of leprosy may become clearer. Valid reasons must be found for the contradictory results from these two major BCG trials, each involving many thousands

of child subjects.

Russell, Scott & Wigley (1964, 1968) and Scott, Wigley & Russell (1966) have also reported a more limited BCG trial started in 1962 in Karimui, eastern New Guinea, among a unique aboriginal population of about 5000 persons of all ages. The prevalence of leprosy is high (60 per 1000) but that of tuberculosis low (3.7% total Mantoux-positive). Leprosy incidence was measured by examinations of the whole population in 1964, 1966 and 1967; 8 cases of leprosy were found in 2318 vaccinated subjects (3.5 per 1000) as against 18 cases in 2295 controls (7.8 per 1000). Russell, Scott & Wigley (1968) stated that no definite conclusions could be reached from the preliminary findings, but the results are encouraging.

"BCG vaccination causes a reduction in incidence in the age-group 10-29 years, but after 30 years of age and under 10 years, no significant differences are noted."

Referring to the findings in mice, Rees (1969b) considered that:

"It is now possible, for the first time to compare the characteristics of *Myco. leprae* isolated in the different parts of the world (Rees, 1965), and to determine whether strain differences exist. This may help to explain the differing protection conferred by BCG vaccine in the three areas."

Nonetheless, the equivocal results of the Uganda, Burma and Karimui trials have not so far confirmed the effectiveness of BCG vaccination against human leprosy. Brown, Stone & Sutherland (1968a) believe that it would be premature to reach such a conclusion at this time.

Myco. leprae infection in mice; implications for epidemiology and control

Solely on the basis of morphological changes and the viability of leprosy bacilli in the footpads of mice, it has been presumed that lepromatous patients become non-infectious after 3 months of minimal sulfone therapy. This finding, which has incalculable implications for leprosy control, still lacks the conclusive proof obtainable only by actual epidemiological observations on human subjects. One such study reported by Worth (1968) therefore requires to be considered in some detail. From the records of out-patient admissions to the Special Skin Clinics in Hong Kong from 1954 to 1960, 64 families were identified in which the index case was a lepromatous parent: (1) with children living during the estimated duration of leprosy prior to therapy, and (2) with additional children born after the same parent had started sulfone therapy but was still bacteriologically positive. The main findings of this retrospective study (reconstructed from the records) are summarized in Table 4.

TABLE 4. Frequency of leprosy in children of families in which one parent with lepromatous leprosy was living at home, by clinical and therapeutic status of parent, during a period of 7-22 years in Hong Kong prior to  $1967^{\circ}$ .

Children observed for 7 years or longer	Age at first exposure (years)	No. of children	No. developing leprosy
Exposed to untreated lepromatous parent	At birth	24	1
(average length of observation: 10.1	1-6	46	9
years)	7–11	25	0
Born after same parent started sulfone therapy,	At birth	30	0
but when still bacteriologically positive	1-6	0	0
(average length of observation: 8.6 years)	7-11	0	
Total		125	10
			1

<sup>a</sup> Data from Worth (1968, pp. 296-302).

The 95 children exposed to untreated lepromatous parents were, of necessity, older and observed for a longer period than were the 30 who were born after the parents had been placed on sulfone therapy. A valid comparison should be limited to the 24 children exposed at birth to untreated parents, with 1 secondary case, and the 30 equally exposed at birth but after therapy was started, with no secondary case. The numbers are obviously small and subject to chance variations, thus making it appear that lepromatous leprosy is less transmissible to children exposed at birth (24 children, 1 case) than to those first exposed at ages of 1-6 years (46 children, 9 cases), whereas leprosy may be presumed to be no longer transmissible to children over the age of 6 years (25 children, no case). The ages at onset of leprosy of the 10 children are not mentioned in the report but it would be unfair, in any event, to include among the 95 children exposed to untreated parents those in whom leprosy developed at 10 years of age or older, inasmuch as few, if any, of the 30 younger children of index cases already treated could have reached 10 years of age when observations ended.

These results were interpreted as the required proof that lepromatous patients become non-infectious almost as soon as dapsone treatment is started. On this basis, Worth (1968) recommended that:

"... all recent household contacts should be examined annually for an indefinite period, except for those persons coming into contact after the start of sulfone therapy in the patient, who will not need intensive surveillance."

In fact, the results of this study are far from conclusive. Clinicians, leprologists and public health administrators responsible for leprosy projects cannot accept the view, on the evidence available up to now, that instant non-infectiousness is achieved in leprosy by dapsone therapy. This assumption has not been borne out by the experience of 20 years of very extensive sulfone therapy all over the world, in which large dosages (by mouse standards) were used.

Final proof of the relationship between the MI and contagiousness in leprosy can come only from a careful and prolonged follow-up of two large groups of children, comparable in all respects but that of exposure to an untreated, or to a treated but still bacteriologically positive, lepromatous parent, i.e., under equal conditions of age at exposure, duration of exposure and length of observation. Such a study would, admittedly, be difficult to set up.

It is worth noting that a retrospective study in Bombay (Figueredo å Balkrishnan, 1967) also attempted to estimate comparative risks of household exposure to treated and untreated bacteriologically positive index cases of leprosy. The subjects of this study were 716 family contacts exposed at birth to lepromatous, "infectious" borderline and "infectious" reactional tuberculoid cases, identified from the patient and contact registers of the Acworth Leprosy Hospital Clinic from 1951 to 1963. Despite limitations in the data, particularly with regard to total numbers of persons at risk (denominator) and to equality between the groups in length of observation, some results of this study (see Table 5) should be considered.

TABLE 5. Leprosy rate in household contacts in Bombay, India, exposed at birth; by therapeutic status of index case, 1951–63<sup>a</sup>

Therapy status of index case	No. observed	Percentage developing leprosy
"Adequately"	190	12.6
Untreated or inade- quately treated	526	29.7
Total	716	25.1

<sup>a</sup> Data from Figueredo & Balkrishnan (1967, pp. 87–96).

<sup>b</sup> Those patients receiving at lease one-third of the prescribed dapsone treatment each year. Administration of sulfones on a voluntary out-patient basis tends to become more irregular in each successive year of treatment but patients would be considered to have taken more than sufficient therapy if judged by mouse footpad standards.

As would be expected, the attack rate is found to be significantly lower for contacts of treated cases (12.6%) than for those of untreated, or inadequately treated, cases (29.7%) but it should be appreciated that a substantial number of leprosy infections (33/190 or 12.6%) presumably still occurred among children exposed at birth to index cases already "adequately" treated.

The Chingleput chemoprophylaxis trial was a strictly supervised, double-blind trial in intrafamilial child contacts of bacteriologically positive and formerly bacteriologically positive leprosy index cases; the later had recently become bacteriologically negative (after dapsone therapy). The children were examined every 3 months and received 92.5% of the prescribed dapsone or matching placebo tablets. The prophylactic doses of dapsone, adjusted to age, were large and approached standard therapeutic levels. For example, children aged 11-14 years received a maximum dose of 300 mg per week from the 4th to the 9th month, followed by a maintenance dose of 150 mg per week from the 10th month onwards. The main findings are summarized in Table 6.

As expected, prophylactic dapsone therapy effected a 53.7% reduction in the incidence of leprosy in this study, as shown by the difference in attack rates between the dapsone group (6.0%) and the controls (13.0%), but it should be noted that an attack rate of 6.0% (19/316) occurred over an average period of 3<sup>1</sup>/<sub>2</sub> years in the dapsone group in children who were, moreover, without any "bacterial load" or signs of leprosy at the start. Allowing for the limitations of the trial, it would even appear that leprosy may develop among child contacts exposed to index cases that are already bacteriologically negative, with or without the benefit of prophylactic dapsone treatment (8/153 or 5.2% against 7/123 or 5.7%).

In conclusion, we repeat that it would be premature to accept the premise that a state of noncontagiousness can be obtained in lepromatous leprosy by 3 months of minimal sulfone therapy, or that BCG vaccination confers definite protection against leprosy, solely on the basis of observed changes in the MI and on the viability of leprosy bacilli in mice. The validity of the laboratory findings on *Myco. leprae* should

	Prophylae	tic dapsone group	Placebo group			
Bacteriological status of index case	Contacts observed	Leprosy cases	Contacts observed	Leprosy cases		
Negative throughout Positive	$\begin{array}{c}153\\163\end{array}$	8 (5.2%) 11 (6.7%)	123 193	7 (5.7%) 34 (17.6%)		
Total	316	19 (6.0%)	316	41 (13.0%)		

TABLE 6. Occurrence of leprosy over a period of  $3\frac{1}{2}$  years in child contacts of the prophylactic dapsone and placebo groups, according to the bacteriological status of the index case, Chingleput trial, Madras, India,  $1962-66^{n}$ .

<sup>s</sup> Data from Dharmendra, Noordeen & Ramanujam (1967).

not be accepted before they are confirmed definitely by acceptable clinico-epidemiological studies in man. If the laboratory findings were applied to human leprosy, present-day measures for treatment and control might be relaxed or reduced to a minimum. Such a policy could have dangerous consequences for the world population already burdened with an estimated 11 million cases of leprosy; in India alone, there could be 2.5 million cases of leprosy, and 300 million inhabitants live in areas where the disease is hyper-endemic.

From changes in the morphology and viability of Mycobacterium leprae in infected mice, some authors have concluded that the majority of smear-postive human lepromatous patients become non-infectious within 3-4 months of the beginning of regular treatment with sulfones, even at dosages equivalent to only one hundredth of the conventional daily dose-namely, 1 mg daily. These very low dosages have, however, not been advocated because of the risk that resistant strains of Myco. leprae might develop. The laboratory findings have already been applied to human leprosy by some leprologists; if they were more widely adopted, leprosy programmes could be seriously affected and the consequences could be serious in endemic areas. Contrary to the results of laboratory tests, the shortcomings of clinical sulfone therapy are evident all over the world from the long times required for lepromatous cases to become bacteriologically negative, from the high

proportion of relapses and from chemoprophylaxis trials in child contacts. Final proof of the relationship between the morphological index, i.e., the proportion of solidly staining bacilli present, and contagiousness can come only from prolonged and well-planned epidemiological studies. It appears that leprosy may develop in appreciable numbers of child contacts exposed to index cases already bacteriologically negative, with or without prophylactic dapsone treatment.

The results of controlled BCG trials now in progress are not consistent although laboratory trials in mice have indicated that BCG vaccination confers a high degree of protection. However, the results that have been obtained in the mouse, an unnatural host, may not be obtainable in man.

It is considered premature to apply laboratory findings to human leprosy before clinical and epidemiological studies have been made in man.

#### RÉSUMÉ

Quelques décourvertes de laboratoire récentes concernant *Mycobacterium leprae*: Leurs conséquences pour le traitement, l'epidemiologie et la lutte antilépreuse.

Se fondant sur des modifications de la morphologie et de la viabilité de *Mycobacterium leprae* constatées chez la souris, certains auteurs ont conclu que la majorité des malades lépromateux à isolements positifs deviennent non contagieux après 3 ou 4 mois d'un traitement régulier par les sulfones, même si la dose utilisée ne représente que le 1/100 de la dose quotidienne classique (100 mg). Quelques léprologues ont déjà mis ces données a profit pour le traitement des malades. Cette pratique, si elle était communément adoptée, pourrait mettre en péril les progrès et l'efficacité des programmes de lutte antilépreuse avec des conséquences peut-être graves pour la population des régions d'endémicité.

Le présent article passe en revue quelques découvertes récentes relatives à Myco. leprae et étudie leurs répercussions en maitière de traitement, d'epidémiologie et de lutte. Chez la souris, le bacille lépreux fait preuve d'une telle sensibilité au dapsone, qu'il suffirait d'une dose minimale d'environ 1 mg par jour pour obtenir une efficacité équivalente chez l'homme. De tels dosages ne sont cependant pas préconisés étant donné le risque d'apparition de souches de Myco, le prae pharmaco-résistantes. Contrastant avec les observations faites chez lo souris, des signes de l'insuffisance du traitement sulfoné se manifestent partout dans le monde, qu'il s'agisse des longs délais nécessaires à la négativation des malades lépromateux, de la proportion élevés des rechutes ou des échecs de la chimiophophylaxie chez les enfants exposés à l'infection. Seule une étude épidémiologique prolongée et soigneusement planifiée pourrait fournir la preuve définitive d'une relation entre l'indice morphologique (proportion des bacilles prenant bien la coloration) et le caractère contagieux de la lèpre. En fait, il semble qu'un nombre appréciable d'enfants en contact avec des malades déjà négativés du point de vue bactériologique sont exposés à contracter la lèpre qu'ils aient ou non bénéficié d'un traitement prophylactique par les sulfones. Les résultats d'essais contrôlés de vaccination par le BCG actuellement en cours sont contradictoires, bien que l'experimentation ait montré que chez la souris le BCG confère un degré élevé de protection.

Les auteurs insistent sur le fait qu'un effet observé chez une espèce animale ne se reproduit pas nécessairement chez l'homme. La souris n'est pas naturellement réceptive à l'infection lépreuse alors que l'homme est un hôte naturel de *Myco. leprae* et peut présenter les manifestations cliniques de la lèpre lépromateuse.

Il se peut que des faits nouveaux viennent corroborer ces découvertes de laboratoire mais il serait prématuré et peu sage d'appliquer ces donnes en léprologie humaine avant qu'elles n'aient été confirmées par des études cliniques et épidémiologiques chez l'homme.

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