LONG-TERM TREATMENT OF DAPSONE-RESISTANT LEPROSY WITH RIFAMPICIN: CLINICAL AND BACTERIOLOGICAL STUDIES

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Although dapsone (DDS) continues to be the standard treatment for the majority of leprosy patients, alternative drugs are required for the therapy of the ever increasing number of patients developing sulphone resistance. Because a high proportion of patients with active lepromatous leprosy in Malaysia now have DDS resistance (mouse foot-pad proven), they have been used in many of our long-term chemotherapeutic trials in leprosy. Clofazimine was the first alternative drug used in our DDS-resistant leprosy trials, but the introduction of rifampicin provided a particularly important alternative drug because of its bactericidal effect with very rapid killing of M.leprae (1,2,3).

Since 1968 we have treated well over 100 lepromatous patients with rifampicin in various studies, including short and longer-term trials. Fifty of these rifampicin treated patients had proven DDS resistance and now some of these patients have been on continuous rifampicin therapy for up to 7 years. Therefore these patients form the basis of our presentation on the long-term treatment of sulphone resistant leprosy with rifampicin. However, because it has been shown that DDS-resistant M.leprae (6 strains) were as sensitive to rifampicin as DDS-sensitive organisms (4 strains) (R.J.W. Rees, personal communication), detailed bacteriological studies were undertaken on 28 of the patients to determine the efficiency of rifampicin to "eradicate" persisting viable M.leprae. We applied the same techniques as we had used for detecting persisting viable M.leprae in lepromatous patients treated for 10 years with DDS (4).

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CLINICAL STUDIES

METHODS. All patients from the start of treatment have been subjected to regular independent clinical, histological (biopsy index) and bacteriological (BI and MI) assessment using our standard methods (5). The incidence and severity of erythema nodosum leprosum (ENL) and the nature and incidence of toxic side effects were also assessed. With one exception rifampicin treatment was 600 mg daily (one patient 600 mg rifampicin once weekly). The majority of patients received combined therapy (rifampicin + thiambutosine, 1.0 g once weekly by injection or 1.0 g twice daily by mouth) in the hope of reducing the emergence of drug resistance; those receiving rifampicin monotherapy were either already known to be thiambutosine/thiacetazone resistance (mouse foot-pad proven) or entered the study before the policy of combined therapy was adopted.

RESULTS. By all assessments all patients showed good and steady improvement irrespective of the particular rifampicin regimen. Initial clinical improvement was particularly rapid being detected in 2-3 weeks (especially relief of nasal blockage) as compared with 2-3 months with DDS. The MI fell just as fast in the DDS resistant patients as in previously untreated lepromatous patients treated with rifampicin. The BI however fell at approximately the same rate as in patients treated with other effective anti-leprosy drugs (Fig. 1). ENL was not significantly more common, severe or rapid in onset than we have experienced in non-resistant lepromatous patients treated with DDS, however the results are not strictly comparable as the resistant patients usually have less severe leprosy than the non-resistant. None of the patients on daily rifampicin have so far developed signs of "immunological" toxicity or produced rifampicin dependent antibodies. However, one patient on 600 mg weekly complained of fever and abdominal symptons after three years of treatment, but without rifampicin dependent antibodies and has now been changed to 600 mg rifampicin daily. There was no decrease in the platelet counts in the patient.

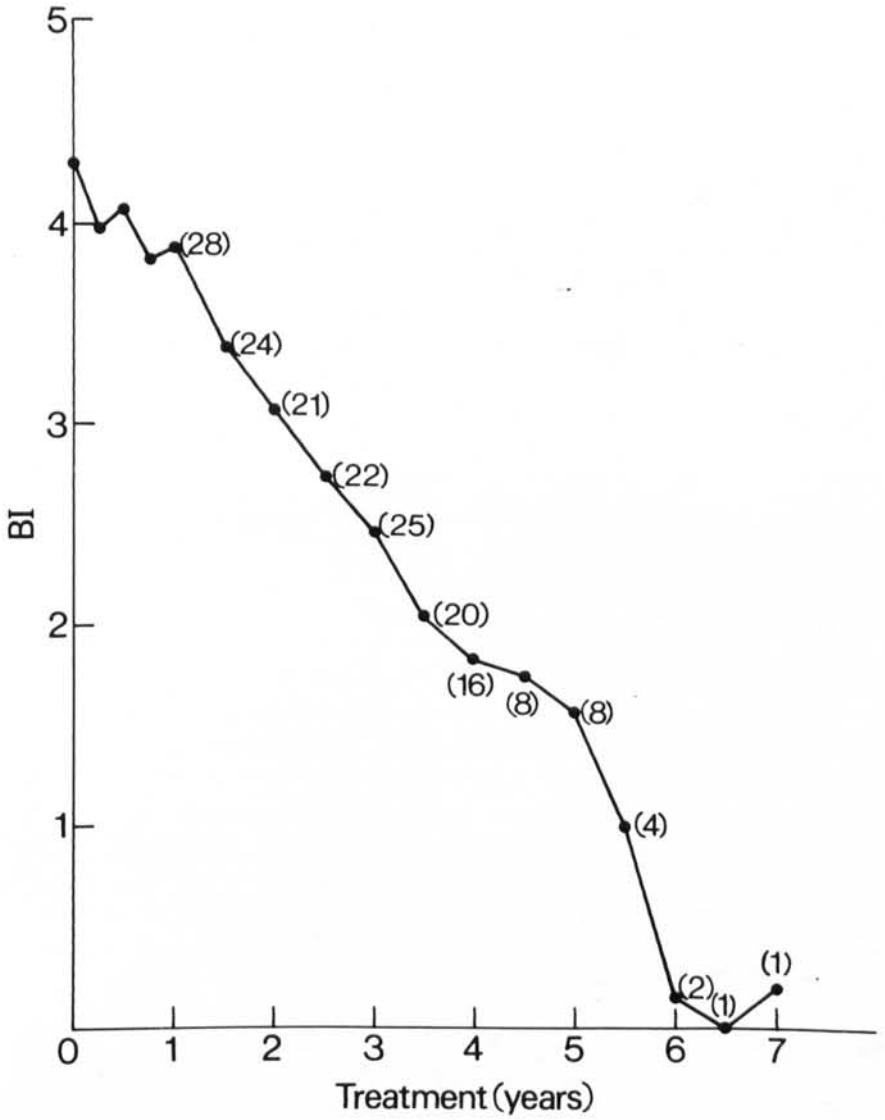

FIG.1. Fall in bacteriological index (BI) in 28 DDS resistant lepromatous patients treated with rifampicin. Number of patients in brackets.

BACTERIOLOGICAL STUDIES

METHODS. In this study the bacteriological progress of the patients was assessed by attempting to isolate viable M.leprae from their tissues by mouse foot-pad inoculation, by the method used for monitoring similarly patients on long-term DDS (4). The tissue biopsies were homogenised, their acid-fast bacillary (AFB) content determined, and the suspension inoculated into both hind foot pads of 6 immunologically suppressed female CBA mice (thymectomy followed by 5 exposures to 200R at 2-week intervals, T/R). Sometimes additional normal mice were also inoculated. The maximum number of AFB inoculated per foot pad was 10^5 for T/R mice and 10^4 for normal mice, however all tissue suspensions were inoculated whether or not the bacillary yields reached these maxima or a countable number of bacilli. Surviving mice were sacrificed 12 months following inoculation and both foot pads harvested individually and an increased yield >3 scored as a positive isolate. Where possible biopsies of skin, nerve (usually superficial radial, occasionally sural), striated muscle (triceps or quadriceps femoris) and scrotal skin (to include smooth dartos muscle) were obtained and each tissue homogenised and processed separately. Twenty-eight of the 50 DDS resistant patients receiving rifampicin regimens were submitted to these special studies and their biopsy tissues monitored for persisting viable M.leprae after 0.5, 1, 2 and 2.5 years treatment, 5 of these patients were monitored a second time, 3-5 years after treatment. The details of these patients are given in Table 1.

RESULTS. Positive isolates from one or more of the biopsied sites were obtained from 4 of 6 patients treated for 0.5 years, 4 of 10 patients treated for 1 year, 9 of 11 patients treated for 2 years and no isolates from the one patient treated for 2.5 years (Table 2). Of the 5 patients monitored a second time, 4 were first monitored at 2 and one at 2.5 years, 3 had positive isolates for the first time at the later monitoring (Table 3). Thus overall, of the 28 patients monitored once or twice after 0.5 to 5 years treat-

TABLE 1

DETAILS OF RIFAMPICIN TREATMENT AND TIMES OF MONITORING *M.LEPRAE* FROM 28 DAPSONE RESISTANT PATIENTS

NUMBER OF PATIENTS			TREATMENT	THIA
TOTAL	RIF + THIA	RIF ALONE	(YEARS)	RESISTANT
6	6[A]	-	0.5	-
10	8	2	1.0	2
11	7	4	2.0	2
1	-	1	2.5	-
28	21	7		4
		MONITORED A SECOND TIME		
3	-	3	3.0	1
1	-	1	4.0	-
1	-	1	5.0	-
5	0	5		1

RIF = Rifampicin: 600 mg daily

THIA = Thiambutosine: 1 g twice daily *per os* or 1 g once weekly by injection

[A] One patient received 600 mg rifampicin once weekly

TABLE 2

PROPORTIONS OF ISOLATES OF M.LEPRAE OBTAINED FROM FOUR TISSUE SITES IN 28 DDS RESISTANT PATIENTS TREATED WITH RIFAMPICIN FOR 0.5 - 2.5 YEARS

TREATMENT (YEARS)	PATIENTS: PROPORTION POSITIVE	BIOPSY SITES PROPORTION POSITIVE			
		SKIN	MUSCLE	NERVE	DARTOS
0.5	4[A]/6	2/6	1/5	1/6	2/2
1.0	4/10	2/9	1/9	1/10	3/8
2.0	9/11	4/11	4/10	4/8	5/6
2.5	0/1	0/1	0/1	-	-

[A]One patient received 600 mg Rifampicin once weekly

TABLE 3

COMPARISON OF ISOLATES OF M.LEPRAE OBTAINED ON TWO OCCASIONS FROM 5 DDS RESISTANT PATIENTS TREATED WITH RIFAMPICIN

PATIENT No.	TREATMENT (YEARS)	BIOPSY SITES POSITIVE (+) OR NEGATIVE (O) SKIN	MUSCLE	NERVE	ISOLATES DARTOS
8859	2.0	O	O	O	-
	3.0	-	-	+	O
12351	2.0	+	+	-	-
	3.0	-	-	+	O
16095	2.0	O	+	O	-
	3.0	-	-	O	O
10743	2.0	O	O	-	-
	4.0	+	-	O	-
10607	2.5	O	O	-	-
	5.0	O	O	+	+

ment with rifampicin, 20 had positive isolates and in particular all 11 patients treated for 2-5 years had positive isolates at one or more of these sites biopsied. There was no significant difference between mono- and combined-therapy in the persistence of viable M.leprae. Positive isolates from 2 patients after 0.5 and one patient after 1 year were tested for rifampicin sensitivity by passage in mice and all proved to be sensitive, all 3 were on combined therapy.

The pattern of results obtained in this study to monitor particular tissues from patients after longer-term rifampicin therapy are similar to those obtained from patients on long-term DDS therapy, based on identical methods. These similarities based now on much more extensive data more firmly justify the pattern as being characteristic of persisting viable M.leprae from lepromatous patients after several years of therapy. Thus, from the four chosen tissues using T/R mice limited multiplication of M.leprae was obtained in mice in 13-25% of the foot pads inoculated. Moreover, at any one time patients harbouring viable bacilli capable of multiplying in the mouse more usually were isolated from one or sometimes two rather than all 4 biopsied sites. There is suggestive evidence that scrotal skin is more likely to harbour such organisms and that from this site they are more likely to multiply in the mouse. Analysis of this data is presented in Table 4.

CONCLUSIONS AND DISCUSSION

Rifampicin treatment is fully effective over a period up to 7 years in the treatment of patients with DDS resistance and this finding is consistent with the results in mice showing that DDS resistant strains of M.leprae are as sensitive to rifampicin as are DDS sensitive strains. Standard clinical, bacteriological and histological assessments showed no evidence of relapse in any of the 50 patients treated with rifampicin and therefore no evidence in the period of time covered of the emergence of rifampicin resistant strains of M.leprae. However, the number of patients involved, the relatively short time covered and the fact that the majority of patients received combined therapy does not exclude the

TABLE 4

DATA ON DISTRIBUTION OF M.LEPRAE ISOLATES FROM THE FOUR BIOPSY SITES IN PATIENTS WITH PERSISTING VIABLE BACILLI

	BIOPSY SITES			
	SKIN	MUSCLE	NERVE	DARTOS
POSITIVE FOOTPAD (%)	13	23	21	25
POSITIVE SITES (%)	47 (9/19)	35 (6/17)	45 (9/20)	73 (11/15)
	POSITIVE SITES			
1 SITE (12 PATIENTS)	2	1	6	3
2 SITES (6 PATIENTS)	4	2	1	5
3 SITES (1 PATIENT)	1	1	0	1
4 SITES (2 PATIENTS)	2	2	2	2

possible emergence of rifampicin resistance. Since the majority of our DDS resistant patients received combined therapy, it is tempting but not conclusive that this regimen prevented the emergence of rifampicin resistance.

We consider that although the patients in this study had "relapsed" active lepromatous leprosy with DDS resistant M.leprae it is justifiable to equate their response to rifampicin to that obtained in previously untreated lepromatous patients. Accepting this assumption the results clearly show that unfortunately our hopes of a quick "cure" by rifampicin have been disappointing, since after 2-5 years of continuous therapy the rifampicin treated patients still harbour persisting viable organisms. Using the same methods we had earlier established the existence of persisting viable M.leprae in previously untreated patients given DDS continuously for 10 years. Since it has been established that such DDS treated patients when taken off treatment can relapse we assume that our criteria for the detection of persisting viable organisms using the mouse fully justifies the method. On this basis therefore we have within a period of 5 years of continuous rifampicin treatment failed to show that this highly bactericidal drug will necessarily eradicate persister organisms any more efficiently than DDS. One must conclude, not with an answer but a question: can patients with lepromatous leprosy ever be cured by chemotherapy alone?

ACKNOWLEDGEMENTS

All the data presented in this paper is part of coordinated chemotherapeutic trials undertaken by the Medical Research Council at the Leprosy Research Unit in Malaysia and more recently in Ethiopia. The trials reported here were coordinated from the National Institute for Medical Research, London, and the clinical conduct and expertise undertaken by Drs J.M.H. Pearson and M.F.R. Waters. The mouse footpad studies were undertaken in London on refrigerated tissues despatched by air. The assessments of rifampicin-dependent antibodies were undertaken by Dr Sheila Worlledge of the Department of Haematology, Royal Postgraduate Medical

School, Hammersmith. Messrs. Gruppo Lepetit of Milan generously provided free all the rifampicin. One of us (RJWR) is grateful to the British Leprosy Relief Association (LEPRA) for providing funds to attend this Conference.

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