THE COURSE OF PULMONARY TUBERCULOSIS IN PATIENTS EXCRETING ORGANISMS WHICH HAVE ACQUIRED RESISTANCE TO ISONIAZID: RESPONSE TO CONTINUED TREATMENT FOR A SECOND YEAR WITH ISONIAZID ALONE OR WITH ISONIAZID PLUS PAS

C. V. RAMAKRISHNAN, A. L. BHATIA, S. DEVADATTA, WALLACE FOX, A. S. L. NARAYANA, J. B. SELKON & S. VELU

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The Course of Pulmonary Tuberculosis in Patients Excreting Organisms which have Acquired Resistance to Isoniazid *

Response to Continued Treatment for a Second Year with Isoniazid Alone or with Isoniazid plus PAS

C. V. RAMAKRISHNAN, A. L. BHATIA, S. DEVADATTA, WALLACE FOX, A. S. L. NARAYANA, J. B. SELKON & S. VELU

This study from the Tuberculosis Chemotherapy Centre, Madras, summarizes the progress during the second year of the patients in a concurrent comparison of four domiciliary chemotherapeutic regimens (isoniazid plus PAS and three regimens of isoniazid alone) who had bacteriologically active or bacteriologically relapsed pulmonary tuberculosis, with isoniazid-resistant organisms, at the end of the first year of treatment.

Of the 57 patients who continued on the same chemotherapy during the second year, nine had attained bacteriological quiescence by the end of that year, 15 still had bacteriologically active disease and 33 had during the year had a serious radiographic deterioration necessitating a change of treatment. An association was observed between the response to treatment in the second year and both the extent of cavitation and the degree of culture-positivity at the end of the first year, but no association was found between the response to treatment in the second year and the level of isoniazid-resistance, the catalase activity, the susceptibility to hydrogen peroxide or the virulence in the guinea-pig of cultures isolated in the last months of the first year.

It is concluded that persisting bacteriological positivity at the end of one year's treatment with isoniazid, either alone or in combination with PAS, is likely to lead to serious radiographic deterioration, irrespective of the level of isoniazid-resistance or of the catalase activity of the cultures of tubercle bacilli, and that, consequently, treatment of patients with bacteriologically active pulmonary tuberculosis must aim at rendering the sputum culture-negative in all instances.

INTRODUCTION

An earlier report from the Tuberculosis Chemotherapy Centre, Madras (Tuberculosis Chemotherapy Centre, 1960) presented the progress in the first year of all the patients in a controlled comparison of isoniazid plus p-aminosalicylic acid (PAS) with three different regimens of isoniazid alone in the domiciliary treatment of pulmonary tuberculosis. It was found that the best treatment in terms of the attainment of bacteriological quiescence was isoniazid plus PAS. A number of authorities believe, however, that bacteriologically active disease with isoniazid-resistant tubercle bacilli is not necessarily a serious state (Deuschle et al., 1954; Middlebrook & Dressler, 1954; Oestreicher et al., 1955; McDermott. 1960). In view of this, as was pointed out in the earlier report (Tuberculosis Chemotherapy Centre, 1960), it was important to study the progress over a

* From the Tuberculosis Chemotherapy Centre, Madras, India. The Centre is under the joint auspices of the Indian Council of Medical Research, the Madras State Government, the World Health Organization and the Medical Research Council of Great Britain.

longer period of the patients who had bacteriologically active disease at 12 months with isoniazid-resistant tubercle bacilli. Further, although two of the isoniazid-alone regimens were clearly unsatisfactory even in the first year, it is only in the light of this longer-term study that it is possible finally to determine the relative value of the third isoniazid-alone regimen (HI-1), a moderate dosage of isoniazid given in one dose a day. The object of the present paper is to report the findings for the second year for all the patients with bacteriologically active or relapsed disease at one year. The progress in the second year of the patients whose disease at one year was bacteriologically quiescent or of doubtful status has been reported elsewhere (Velu et al., 1961).

GENERAL PLAN AND CONDUCT OF THE STUDY

The patients who had bacteriologically active or relapsed disease (as defined below) at the end of one year of the initially prescribed chemotherapy continued on the same regimen for the second year unless a serious clinical or clear-cut radiographic deterioration occurred. In this way it was hoped to discover whether continued chemotherapy with the same drug or combination of drugs would lead to quiescence of the disease or whether it was necessary in such patients to change the chemotherapy-a point of considerable importance in planning large-scale campaigns against tuberculosis.

THE CHEMOTHERAPY

The four regimens studied in the first year and prescribed for the second year for the patients with bacteriologically active or relapsed disease were:

PH. Isoniazid 3.9-5.5 mg/kg body-weight, plus PAS (sodium) 0.2-0.3 g/kg, daily, divided into two doses, by mouth–i.e., 200 mg of isoniazid plus 10 g of PAS (sodium) a day for a patient weighing 100 lb. *(45.4* kg).

HI-1. Isoniazid alone, 7.8-9.6 mg/kg daily, in one dose by mouth–i.e., 400 mg of isoniazid a day for a patient weighing 100 lb.

HI-2. Isoniazid alone, 7.8-9.6 mg/kg daily, divided into two doses, by mouth–i.e., 400 mg of isoniazid a day for a patient weighing 100 lb.

H. Isoniazid alone, 3.9-5.5 mg/kg daily, divided into two doses, by mouth–i.e., 200 mg of isoniazid a day for patient weighing 100 lb.

The precise dosage scales have been detailed in the earlier report (Tuberculosis Chemotherapy Centre, 1960). All the patients were treated at home and were encouraged to remain fully ambulant.

DEFINITION OF BACTERIOLOGICALLY ACTIVE DISEASE AT ONE YEAR

A patient was considered to have bacteriologically active disease at one year if the cultures were never all negative at three consecutive monthly examinations during the 12-month period (the patients had, on the average, nearly three specimens cultured each month).

DEFINITION OF BACTERIOLOGICALLY RELAPSED DISEASE AT ONE YEAR

A patient was considered to have bacteriologically relapsed disease at one year if two or more positive cultures were obtained at the last three monthly examinations (that is, at 10, 11 and 12 months) after a period of three or more consecutive months during which all the cultures were negative.

NUMBER OF PATIENTS IN THE SECOND-YEAR STUDY

At the end of one year's treatment (Table 1) 61 patients who were on the initially prescribed chemo therapy were classified as having bacteriologically active or relapsed disease-namely, six (7%) of 86 PH patients, 12 (19%) of 64 HI-1, 13 (20%) of 66 HI-2 and 30 (35 %) of 86 H patients. In addition, by that time, two (2%) of the PH patients, five (8%) of the HI-1 patients, 15 (23 %) of the HI-2 patients and 18 (21%) of the H patients had either died of tuberculosis or had had their chemotherapy changed during the first year of treatment because of serious radiographic or clinical deterioration–evidence that, even in the first year, the HI-2 and H regimens were unsatisfactory. Of the 61 patients who had active or relapsed disease at one year, three (1 PH, 2 HI-2) did not continue on the initially prescribed chemotherapy in the second year as they had been incorrectly classified at the end of the year's treatment on the basis of the then available

TABLE 1
CLASSIFICATION OF THE PATIENTS AT ONE YEAR ACCORDING TO THEIR RESPONSE TO TREATMENT

Classification at one year		Treatment series							
		PH		HI-1		HI-2		H	
		No.	%	No.	%	No.	%	No.	%
Favourable response	Bacteriologically quiescent disease	74	86	43	67	37	56	38	44
	Disease of bacteriologically doubtful status	4	5	4	6	1	2	0	0
Unfavourable response	Bacteriologically relapsed disease	4	5	2	3	5	8	5	6
	Bacteriologically active disease *without* radiographic deterioration	2	2	10	16	8	72	25	29
	Bacteriologically active disease with deterioration necessitating a change of chemotherapy	1	1	5	8	14	21	15	17
	Tuberculous death	1	1	0	0	1	2	3	3
	Total patients	86	100	64	100	66	101	86	99

bacteriological data as having disease that was bacteriologically quiescent or of doubtful status (all the culture results were not available at that time). A further patient (H), who had radiographically static tuberculous disease and who also had severe emphysema at 12 months, had his treatment changed in the thirteenth month on account of cor pulmonale. There remained 57 patients (5 PH, 12 HI-1, 11 HI-2, 29 H) who had active or relapsed disease at the end of the first year and continued on the initially prescribed chemotherapy in the second year.

GENERAL MANAGEMENT AND ROUTINE INVESTIGATIONS

During the second year, the patients attended the Centre weekly or fortnightly for a supply of their medicine, and monthly for a routine examination. This examination included a full-plate postero-anterior chest radiograph and the examination of a minimum of one overnight collection specimen of sputum by smear and culture, or, if no sputum was being produced, the examination of a pair of laryngeal swabs by culture, each on a separate slope; the two swabs were regarded as a single specimen. A test of sensitivity to isoniazid was set up on one positive culture at each month.

COLLAPSE THERAPY AND RESECTION

None of the patients had any form of collapse therapy or resection during the first or the second year.

INDEPENDENT ASSESSMENT OF THE RADIOGRAPHS

Radiographic assessments of the severity of the disease at 12 months were made from full-plate postero-anterior radiographs (tomographs were used for the assessment of the extent of cavitation) by an independent assessor (Dr Raj Narain) who was unaware of the treatment the patients had received. The assessments were: the extent of cavitation, graded as extensive, moderate, slight or nil; the total extent of the radiographic lesions, classified as gross, extensive, moderate, limited, slight, trivial or nil (Tuberculosis Chemotherapy Centre, 1960); and the number of lung zones involved in disease, standard definitions of zones (Daniels et al., 1948) being used and the presence of any disease in each zone, no matter how limited in extent, being recorded.

If, during the course of the second year, a patient was considered by the Centre's medical staff to have a definite radiographic extension of the disease, the following procedure was adopted. The supervision of the patient was intensified. A 7- to 10-day course of penicillin was given and a further radiograph was taken at the end of two weeks or at the next monthly examination. Careful supervision for evidence of radiographic evolution of the lesion was maintained. If the new lesion was persisting or spreading, and was more than minor, the complete radiographic series was shown to an independent assessor (Dr K. S. Sanjivi) who was unaware of the treatment the patient was receiving. This procedure was adopted because of the observation of

Oestreicher et al. (1955) that the radiographic spreads which occur under treatment with isoniazid alone frequently resolve without a change of chemotherapy. The assessor sometimes decided that the disease had deteriorated radiographically to such an extent that chemotherapy should be changed. In other instances, he considered a change of chemotherapy was not warranted and indicated if and when he wished to see the radiographic series again. In addition, serious clinical deterioration was regarded as a reason for changing the chemotherapy without reference to an independent assessor. At the end of two years the full radiographic series of all the patients who continued on the initially prescribed regimen until that time and who still had bacteriologically active disease were reviewed by the independent assessor to ensure that all the radiographic spreads warranting a change of treatment had been detected.

DEFINITION OF BACTERIOLOGICAL QUIESCENCE AT TWO YEARS

A patient's disease was classified as bacteriologically quiescent at the end of the second year if not more than one of the specimens examined at the last six monthly examinations was positive on culture.

BACTERIOLOGICAL METHODS

Examination of sputum specimens and laryngeal swabs ; sensitivity tests

The methods used for examining sputum specimens and laryngeal swabs and for determining the sensitivity of cultures to isoniazid have been described in detail elsewhere (Tuberculosis Chemotherapy Centre, 1959, 1960). In brief, tests of sensitivity to isoniazid were performed by inoculating a 3-mm diameter loopful of a 4 mg/ml (moist weight) aqueous suspension of growth from a Löwenstein-Jensen medium culture on to a slope of this medium without isoniazid and on to a series of slopes containing 0.2, 1, 5 and 50 µg/ml of isoniazid. The degree of growth was recorded after four weeks' incubation at 37°C. The minimal inhibitory concentration (MIC) of isoniazid was defined in two ways-namely, as the lowest concentration of isoniazid that prevented the growth of 20 colonies or more (the 20-colony MIC) and as the lowest concentration of isoniazid that prevented an equal amount of growth to that on the drug-free control slope (the equal-growth MIC). The patients were classified according to the level of resistance to isoniazid of the majority of the cultures isolated at 10, 11 and 12 months which were tested for their sensitivity to isoniazid. If no majority could be obtained, as, for example, when there were only two cultures and they gave different results, then the result of the culture isolated at nine months was included. As shown in Table 2, patients were classified according to the 20-colony definition as having harboured organisms with " very high " resistance if the majority of the cultures had MICs which were greater than 50 µg/ml, with "high " resistance if the majority of the MICs were 50 µg/ml, with " moderate " resistance if the majority of the MICs were 5 µg/ml and with " low " resistance if the majority of the MICs were 1 µg/ml. The classification of the patients according to the equal-growth MIC was the same (Table 2), but included an additional category of " sensitive " for those patients who had a majority of cultures with MICs of 0.2 µg/ml or less-that is, where the growth on 0.2 µg/ml was less than that on the control slope.

Catalase activity

The catalase activity of the cultures tested for their sensitivity to isoniazid was determined by the semiquantitative method described by Kreis, Le Joubioux & Pariente (1956). In brief, a suspension of the growth (approximately 4 mg moist weight) was exposed to contact with a 0.3 % (w/v) aqueous solution of hydrogen peroxide at pH 7 for one hour at 8°C. After the addition of 0.5 ml of N sulfuric acid the residual peroxide was estimated by titration with potassium permanganate. The catalase activity was expressed as the percentage of the peroxide destroyed (to the nearest 10%).

Susceptibility to hydrogen peroxide

The susceptibility of the cultures to hydrogen peroxide was determined by the method described by Subbaiah, Mitchison & Selkon (1960). In brief, cultures were exposed to 0.02% (w/v) hydrogen peroxide in pH 7 buffer solution for 90 minutes and the proportion of the population which survived was determined by viable counting. The susceptibility to hydrogen peroxide was expressed as the proportion of the population of organisms tested which survived this exposure.

Virulence of tubercle bacilli in the guinea-pig

The virulence of cultures of tubercle bacilli in the guinea-pig was measured by inoculating intra-

TABLE 2

CLASSIFICATION OF THE PATIENTS ACCORDING TO THE LEVEL OF RESISTANCE TO ISONIAZID OF THE CULTURES ISOLATED AT 10, 11 AND 12 MONTHS

Classification of the level of isoniazid-resistance	20-colony MIC [a] of isoniazid (µg/ml) [b]					Equal-growth MIC [a] of isoniazid (µg/ml)					
	Number of patients	1	5	50	> 5 0	Number of patients	< 0.2	1	5	50	> 50
Very high	10				3	4					3
	3				2	2					2
	4				1 [c]	2				1	2
	6			1	2 [d]	2			1		2
						1	1				2
High	6			3		6				3	
	2			2		1				2	
	3			2	1	2				1 [c]	
	8		1	2 [d]		1				2	1
						2			1	2	
						1		1		2 [d]	
						2	1			2 [d]	
						1			1	1 [c]	1
						2		1		1	1
Moderate	5		3			7			3		
	2		2			3			2		
	2		1			2			1		
	2		2	1		1		1	2		
	1	1	2			2		1	1	1 [c]	
						1		1	1		1
Low	2	3				3		3			
	1	2				1		1			
						2		2	1		
						1		2		1	
" Sensitive "						1	2				
						2	1				
						2	2				

[a] Minimal inhibitory concentration.
[b] The figures in each column under the MIC denote the number of culture results per patient.
[c] 9-month culture result.
[d] Including a 9-month culture result for one patient.

muscularly 1.0 mg (moist weight) of the culture into each of two guinea-pigs, one of which was killed at 6 weeks and the other at 12 weeks, unless it had already died. At post-mortem examination, the total extent of tuberculous disease in the spleen, liver, lungs and local glands was assessed as a score ranging from 0 to 100. The square-root of the ratio of the score to the survival period in days was determined for each guinea-pig (whether sacrificed or dead from tuberculosis), and was termed the

root-index. The measure of virulence employed was the mean of the root-indices for the two guinea-pigs inoculated with each culture, and was termed the root-index of virulence (Mitchison et al., 1961). The virulence tests were done in two series of experiments, the larger at the Microbiological Research Establishment, Porton, England, and the smaller at the Centre in Madras; the results of the Madras series were adjusted to those obtained at Porton as described by Mitchison et al. (1962).

RESULTS

The progress during the second year of the 61 patients who were classified as having had bacteriologically active or relapsed disease at 12 months will be presented in two sub-sections. The first reports on the 57 patients who continued on the originally prescribed chemotherapy in the second year and the second on the three patients who, in error, had their chemotherapy changed at one year and on the patient who had his treatment changed on account of cor pulmonale.

PROGRESS IN THE SECOND YEAR OF THE PATIENTS WHO CONTINUED ON THE ORIGINALLY PRESCRIBED CHEMOTHERAPY

Response to treatment

The progress in the second year of the 57 patients with bacteriologically active or bacteriologically relapsed disease at one year who continued on the originally prescribed chemotherapy is set out in Table 3. One (HI-2) of the 57 patients insisted on having his treatment changed at 18 months, at which time he still had consistently positive cultures with radiographically static disease. Despite a change to streptomycin plus PAS his cultures remained positive up to and including 21 months. This patient has been classified in Table 3 and in all the analyses as having active disease without radiographic deterioration at two years. Bacteriological quiescence was achieved in nine of the 57 patients-namely, one of five PH patients, two of 12 HI-1, one of 11 HI-2 and five of 29 H patients. The month at which the last positive culture was obtained was the twelfth month for the PH patient, the tenth month for one of the HI-1 patients and the eleventh month for the other (apart from isolated positive cultures at 15 and 24 months), the twelfth month for the HI-2 patient (apart from an isolated positive culture with l-colony growth at 17 months) and the tenth, tenth, eleventh, eleventh and thirteenth months, respectively, for the five H patients. Thus, all except three of the patients who attained bacteriological quiescence during the second year had already converted their sputum to bacteriological negativity late in the first year; and the exceptions did so very early in the second year. All the patients who were bacteriologically positive at 14 months either had active disease for the rest of the period or else deteriorated radiographically to an extent that warranted a change of treatment. Fifteen patients (1 PH, 3 HI-1, 6 HI-2, 5 H) had active disease throughout the second year but without definite radiographic deterioration (Table 3). Radiographic deterioration, warranting a change of treatment, occurred in 33 patients (3 PH, 7 HI-1, 4 HI-2, 19 H). The month in which the deterioration occurred ranged from the thirteenth to the twenty-fourth (Table 3).

Bacteriologically quiescent disease was attained by the end of the second year by five (8%) of the 43 patients classified as having active disease at one year, and by four (35%) of the 14 patients with relapsed disease at one year. The proportions of patients whose disease deteriorated in the second year was similar for those with active and with relapsed disease at one year.

In summary, of the 57 patients, nine (16%) attained bacteriological quiescence, 15 (26 %) still had active disease at two years and 33 (58%) had deteriorated radiographically to an extent which warranted a change of treatment. There were no important differences between the response to treatment in the four treatment series.

Sensitivity to isoniazid of cultures isolated during the second year

Tests of sensitivity to isoniazid were done on 257 of the positive cultures isolated from the 57 patients during the second year. Of these cultures, 255 were resistant to isoniazid (growth of 20 colonies or more on 0.2 μg/ml or a higher concentration) and two (0.8 %) were sensitive. These two cultures were from a PH and an H patient and were isolated sensitive cultures among a series of six and seven resistant cultures, respectively. These observations confirm

TABLE 3

RESPONSE TO TREATMENT IN THE SECOND YEAR OF THE PATIENTS WHO HAD BACTERIOLOGICALLY ACTIVE OR RELAPSED DISEASE AT ONE YEAR AND WHO CONTINUED ON THE INITIALLY PRESCRIBED CHEMOTHERAPY

Treatment series	Disease status at one year	Number of patients	Status of the disease at two years				
			Bacteriologically quiescent		Bacteriologically active	Radiographic deterioration warranting a change of treatment	
			Number	Month at which last positive culture was obtained		Number	Month of deterioration
PH	Active	2	0	—	0	2	16, 24
	Relapsed	3	1	12	1	1	21
	Total	5	1	–	1	3	–
HI-1	Active	10	1	10	3	6	13, 15, 18, 18, 18, 20
	Relapsed	2	1	11	0	1	20
	Total	12	2	–	3	7	—
HI-2	Active	7	0	-	5	2	14, 21
	Relapsed	4	1	12	1	2	18, 21
	Total	11	1	–	6	4	–
H	Active	24	4	10, 10, 11, 13	5	15	13, 13, 13, 15, 16, 16, 17, 18, 18, 19, 19, 19, 20, 21, 23
	Relapsed	5	1	11	0	4	13, 16, 17, 24
	Total	29	5	–	5	19	–
All series	Active	43	5	–	13	25	–
	Relapsed	14	4	–	2	8	–
	Total patients	57	9	–	15	33	—

that the strains which the patients were excreting were consistently isoniazid-resistant.

Response in the second year related to radiographic and bacteriological assessments at the end of the first year

Extent of cavitation. Bacteriologically quiescent disease was attained by 23 % of 35 patients with no or slight cavitation at 12 months, but by only 5 % of 22 patients with moderate or extensive cavitation at one year (Table 4). However, this difference fails to attain significance (P = 0.07). Deterioration warranting a change of treatment occurred in 33% of 21 patients without cavitation at 12 months, as compared with 72% of 36 who had cavitation, a significant difference (P = 0.01). Considering only the patients whose disease did not attain bacteriological quiescence, deterioration occurred in 47 % of 15 patients with no cavitation and in 79% of 33 with cavitation at one year, a difference that also attains. significance (P = 0.05).

Number of lung zones involved in disease. Bacteriologically quiescent disease was attained by 27% of 22 patients with two or fewer lung zones involved in disease at 12 months, as compared with 9% of 35 patients with three or more lung zones involved; while 50% of 22 patients with two or fewer lung zones involved in disease deteriorated to an extent which warranted a change of treatment, as compared with

TABLE 4

RESPONSE TO TREATMENT IN THE SECOND YEAR RELATED TO THE EXTENT OF CAVITATION AT ONE YEAR

Extent of cavitation at one year	Treatment series	Patients	Status of the disease at two years: Bacteriologically quiescent	Bacteriologically active	Radiographic deterioration warranting a change of treatment
		No. %	No. %	No. %	No. %
Nil	PH	2	1	1	0
	HI-1	3	1	1	1
	HI-2	8	1	4	3
	H	6	3	2	3
	All series	21 *100*	6 *(29)*[a]	8 *(38)*	7 *(33)*
Slight	PH	1	0	0	1
	HI-1	2	0	0	2
	HI-2	1	0	0	1
	H	10	2	2	6
	All series	14 *9 9*	2 *(14)*	2 *(14)*	10 *(71)*
Moderate	PH	2	0	0	2
	HI-1	5	1	2	2
	HI-2	2	0	2	0
	H	9	0	1	8
	All series	18 *101*	1 *(6)*	5 *(28)*	12 *(67)*
Extensive	PH	0	0	0	0
	HI-1	2	0	0	2
	HI-2	0	0	0	0
	H	2	0	0	2
	All series	4 *100*	0 *(0)*	0 *(0)*	4 *(100)*
Total patients		57 *100*	9 *16*	15 *26*	33 *58*

[a] The parentheses indicate percentages based on fewer than 25 observations.

TABLE 5

RESPONSE TO TREATMENT IN THE SECOND YEAR RELATED TO THE NUMBER OF LUNG ZONES INVOLVED IN DISEASE AT ONE YEAR

Number of lung zones involved in disease at one year	Treatment series	Patients	Status of the disease at two years: Bacteriologically quiescent	Bacteriologically active	Radiographic deterioration warranting a change of treatment
		No. %	No. %	No. %	No. %
0, 1 or 2	PH	4	1	1	2
	HI-I	2	1	0	1
	HI-2	6	1	2	3
	H	10	3	2	5
	II series	22 *100*	6 (27)[a]	5 *(23)*	11 *(50)*
3	PH	1	0	0	1
	HI-1	6	1	2	3
	HI-2	2	0	1	1
	H	9	0	2	7
	II series	18 *101*	1 *(6)*	5 *(28)*	12 *(67)*
4 or 5	PH	0	0	0	0
	HI-1	4	0	1	3
	HI-2	3	0	3	0
	H	10	2	1	7
	II series	17 *100*	2 *(12)*	5 *(29)*	10 *(59)*
Total patients		57 *100*	9 *16*	15 *26*	33 *58*

[a] The parentheses indicate percentages based on fewer than 25 observations.

63 % of 35 with three or more lung zones involved (Table 5). However, neither of these differences attains statistical significance.

Total extent of disease. There was no clear evidence of any association between the status of the disease at two years and the over-all radiographic extent of the disease at 12 months (Table 6). However, it will be noted that none of the four patients whose extent of disease at 12 months was classified as slight, trivial or nil deteriorated during the second year, as compared with 61% of 33 patients with limited disease and 65 % of 20 patients with moderate or extensive disease.

Degree of culture-positivity. The patients were classified, on the basis of a scoring system, according to the degree of growth on culture of all their sputum and laryngeal swab specimens examined at 10, 11

TABLE 6

RESPONSE TO TREATMENT IN THE SECOND YEAR RELATED TO THE TOTAL EXTENT OF DISEASE AT ONE YEAR

Total extent of disease at one year	Treatment series	Patients	Status of the disease at two years: Bacteriologically quiescent	Bacteriologically	Radiographic deterioration warranting a change of treatment
		No. %	No. %	No. %	No. %
Nil, trivial or slight	PH	1	0	1	0
	HI-1	0	0	0	0
	HI-2	1	0	1	0
	H	2	2	0	0
	All series	4 100	2 (50)[a]	2 (50)	0 (0)
Limited	PH	1	0	0	1
	HI-1	5	1	1	3
	HI-2	9	1	4	4
	H	18	2	4	12
	All series	13 100	4 12	9 27	20 6 1
Moderate or extensive	PH	3	1	0	2
	HI-1	7	1	2	4
	HI-2	1	0	1	0
	H	9	1	1	7
	All series	20 100	3 (15)	4 (20)	13 (65)
Total patients . . .		17 100	9 1 6	15 26	33 5 8

[a] The parentheses indicate percentages based on fewer than 25 observations.

and 12 months (an average of seven to nine cultures). A score of 4 was given for a 3-plus positive culture (confluent growth), 3 for a 2-plus positive culture (innumerable discrete colonies), 2 for a 1-plus positive culture (100-20 colonies), 1 for the presence of 19 to 1 colonies and 0 for a negative culture. The mean score for each patient was calculated and the findings are set out in Table 7. Of the 17 patients who had mean scores of less than 1.0, 47 % attained bacteriological quiescence during the second year, as compared with 2% of the 40 patients who had a mean score of 1.0 or more, a highly significant difference ($P < 0.001$). Conversely, only 29% of the

TABLE 7

RESPONSE TO TREATMENT IN THE SECOND YEAR RELATED TO THE MEAN SCORE FOR CULTURE-POSITIVITY AT 10, 11 AND 12 MONTHS

Mean score for culture-positivity at 10, 11 and 12 months	Treatment series	Patient	Status of the disease at two years: Bacteriologically quiescent	Bacteriologically active	Radiographic deterioration warranting a change of treatment
		No. %	No. %	No. %	No. %
0.0-	PH	0	0	0	0
	HI-I	2	2	0	0
	HI-2	3	1	1	1
	H	4	3	0	1
	All series	9 100	6 (67)[a]	1 (71)	2 (22)
0.5-	PH	2	1	1	0
	HI-1	0	0	0	0
	HI-2	1	0	0	1
	H	5	1	2	2
	All series	8 101	2 (25)	3 (38)	3 (38)
1.0-	PH	1	0	0	1
	HI-1	3	0	0	3
	HI-2	2	0	1	1
	H	1	0	0	1
	All series	7 100	0 (0)	1 (14)	6 (86)
1.5-	PH	0	0	0	0
	HI-I	4	0	2	2
	HI-2	1	0	1	0
	H	2	1	0	1
	All series	7 100	1 (14)	3 (43)	3 (43)
2.0-	PH	1	0	0	1
	HI-1	3	0	1	2
	HI-2	1	0	0	1
	H	10	0	2	8
	All series	15 100	0 (0)	3 (20)	12 (80)
2.5-2.9	PH	1	0	0	1
	HI-1	0	0	0	0
	HI-2	3	0	3	0
	H	7	0	1	6[b]
	All series	11 100	0 (0)	4 (36)	7 (64)
Total patients		57 100	9 16	15 2 6	33 5 8
Mean score for culture-positivity			0.5	1.8	1.8

[a] The parentheses indicate percentages based on fewer than 25 observations.

[b] Including one patient with a mean score of 3.1.

17 patients who had mean scores of less than 1.0 deteriorated to an extent that warranted a change of treatment, as compared with 70% of the 40 patients who had mean scores of 1.0 or more; this difference also attains significance (P = 0.01). This finding that the response to treatment in the second year was related to the degree of culture-positivity in the last three months of the first year must be interpreted with caution, for it will be appreciated that, as already mentioned (page 6), 67% of the nine patients who attained bacteriological quiescence had already started to yield negative cultures before the end of the first year. Furthermore, considering only the patients who did not attain quiescence by the end of the second year, 56% of nine patients with positivity scores of less than 1.0 deteriorated to an extent that warranted a change of treatment, as compared with 72% of 39 with scores of 1.0 or more, a difference which does not attain statistical significance.

Since there was an association between the attainment of bacteriological quiescence by the end of the second year and both the extent of cavitation and the degree of culture-positivity at the end of the first year, the prognostic value of a combination of these two assessments was studied. Of the 12 patients without cavitation who had culture-positivity scores of less than 1.0, six attained quiescence, three remained bacteriologically active and three deteriorated during the second year. Thus, a combination of these two assessments was of no greater prognostic value than culture-positivity alone.

In summary, the response of the patients during the second year was related to the extent of cavitation and to the degree of culture-positivity, but only to a minor extent, if at all, to the number of lung zones involved or the total extent of disease at the end of the first year.

Level of isoniazid-resistance :

(a) 20-colony MIC. Patients were classified (see page 4) according to the level of isoniazid-resistance (20-colony definition) of the cultures at 10, 11 and 12 months. This classification is related to the response to treatment in the second year in Table 8. Of the 42 patients with cultures of very high or high resistance to isoniazid, 14 % attained bacteriological quiescence, as compared with 25% of 12 patients with cultures of moderate resistance and none of three with cultures of low resistance. There was a suggestion that a larger proportion of deteriorations occurred in the patients with cultures of high or very high resistance (64% of 42) than in patients with cultures of moderate or low resistance (40% of 15). However, this difference does not attain significance (P ~ 0.2). Thus, there was little evidence that pro-

TABLE 8

RESPONSE TO TREATMENT IN THE SECOND YEAR RELATED TO THE LEVEL OF ISONIAZID-RESISTANCE AT THE END OF THE FIRST YEAR, ACCORDING TO THE 20-COLONY MIC

Level of isoniazid-resistance (20-colony MIC) at the end of the first year [a]	Treatment series	Patients	Status of the disease at two years		
			Bacteriologically quiescent	Bacteriologically	Radiographic deterioration warranting a change of treatment
		No. %	No. %	No. %	No. %
Very high	PH	1	1	0	0
	HI-1	3	1	1	1
	HI-2	7	1	4	2
	H	12	3	1	8
	II series	23 100	6 (26)[b]	6 (26)	11 (48)
High	PH	1	0	0	1
	HI-I	5	0	1	4
	HI-2	3	0	1	2
	H	10	0	1	9
	II series	19 100	0 (0)	3 (16)	16 (84)
Moderate	PH	2	0	1	1
	HI-I	3	1	0	2
	HI-2	1	0	1	0
	H	6	2	2	2
	All series	12 100	3 (25)	4 (33)	5 (42)
Low	PH	1	0	0	1
	HI-1	1	0	1	0
	HI-2	0	0	0	0
	H	1	0	1	0
	All series	3 100	0 (0)	2 (67)	1 (33)
Total patients . . .		57 100	9 16	15 26	33 58

[a] For definitions, see page 4.

[b] The parentheses indicate percentages based on fewer than 25 observations.

gress during the second year was related to the level of isoniazid-resistance (20-colony definition) at the end of the first year of treatment.

(b) Equal-growth MIC. Table 9 relates the response to treatment in the second year to the classification of the patients according to the equal-growth definition of isoniazid-resistance. Considering the 45 patients with cultures of moderate or higher degree of resistance, 18% had quiescent diease at two years, 18 % remained bacteriologically active and 64 % had deteriorated to an extent that warranted a change of treatment. In comparison, of the 12 patients with low resistance or " sensitive " cultures, the corresponding figures were 8%, 58% and 33%, respectively. Thus there was little evidence that the progress of the patients during the second year was related to the level of resistance (equal-growth definition) of the cultures isolated at the end of the first year.

It remained a possibility that an association between the response to treatment in the second year and the level of resistance to isoniazid at one year was present, but that it was obscured in the above analyses by the other prognostic factors. Analyses (not tabulated here) were therefore undertaken to examine the relationship between response to treatment and the level of resistance according to both the 20-colony and the equal-growth definitions of the MIC, after stratifying the patients according to the extent of cavitation and according to the degree of culture positivity at the end of the first year. No clear associations between response to treatment and the level of resistance were observed.

Catalase activity. Table 10 relates the response to treatment in the second year to the mean catalase activity (semiquantitative method) of the cultures isolated during the last three months of the first year. There were 17 patients with strains with no, or very low, catalase activity (0-19%) and of these 24% attained bacteriological quiescence, 24% remained bacteriologically active and 53 % had a deterioration warranting a change of treatment. There were 15 patients whose strains had a catalase activity of 20-29 %, and of these 13 % attained bacteriological quiescence, 33 % remained bacteriologically active and 53 % had a deterioration warranting a change of treatment. Considering catalase activity of 30-49 %, 27% of 11 patients attained bacteriological quiescence, 9 % remained bacteriologically active and 64 % deteriorated. Of the 14 patients with strains of high catalase activity (50% or more), none attained bacteriological quiescence, 36 % remained bacterio-

TABLE 9

RESPONSE TO TREATMENT IN THE SECOND YEAR RELATED TO THE LEVEL OF ISONIAZID-RESISTANCE AT THE END OF THE FIRST YEAR, ACCORDING TO THE EIDUAL-GROWTH MIC

Level of isoniazid-resistance (equal-growth MIC) at the end of the first year [a]	Treatment series	Patients	Status of the disease at two years		
			Bacteriologically quiescent	Bacteriologically active	Radiographic deterioration warranting a change of treatment
		No. %	No. %	No. %	No. %
Very high	PH	1	1	0	0
	HI-1	2	0	1	1
	HI-2	2	0	1	1
	H	6	1	1	4
	All series	11 100	2 (18)[b]	3 (27)	6 (55)
High	PH	0	0	0	0
	HI-1	3	0	1	2
	HI-2	3	1	0	2
	H	12	2	1	9
	All series	18 100	3 (17)	2 (11)	13 (72)
Moderate	PH	2	0	1	1
	HI-I	5	2	0	3
	HI-2	3	0	2	1
	H	6	1	0	5
	A II series	16 100	3 (19)	3 (19)	10 (62)
Low	PH	1	0	0	1
	HI-1	1	0	0	1
	HI-2	2	0	2	0
	H	3	0	2	1
	A II series	7 100	0 (0)	4 (57)	3 (43)
"Sensitive'	PH	1	0	0	1
	HI-1	1	0	1	0
	HI-2	1	0	1	0
	H	2	1	1	0
	A II series	5 100	1 (20)	3 (60)	1 (20)
Total patients . . .		57 100	9 16	15 26	33 58

[a] For definitions, see page 4.

[b] The parentheses indicate percentages based on fewer than 25 observations.

TABLE IO

RESPONSE TO TREATMENT IN THE SECOND YEAR RELATED TO THE MEAN CATALASE ACTIVITY OF THE CULTURES AT 10, 11 AND 12 MONTHS

Mean catalase activity at 10, 11 and 12 months (%)	Treatment series	Patients	Status of the disease at two years: Bacteriologically quiescent	Status of the disease at two years: Bacteriologically active	Status of the disease at two years: Radiographic deterioration warranting a change of treatment
		No. %	No. %	No. %	No. %
0-19	PH	0	0	0	0
	HI-1	2	0	1	1
	HI-2	5	1	2	2
	H	10	3	1	6
	All series	17 *101*	4 *(24)*[a]	4 *(24)*	9 *(53)*
20-29	PH	3	1	1	1
	HI-1	2	0	1	1
	HI-2	3	0	2	1
	H	7	1	1	5
	All series	15 *9 9*	2 *(13)*	5 *(33)*	8 *(53)*
30-49	PH	1	0	0	1
	HI-1	5	2	0	3
	HI-2	1	0	1	0
	H	4	1	0	3
	All series	11 *100*	3 *(27)*	1 *(9)*	7 *(64)*
50 or more	PH	1	0	0	1
	HI-1	3	0	1	2
	HI-2	2	0	1	1
	H	8	0	3	5
	All series	14 *100*	0 *(0)*	5 *(36)*	9 *(64)*
Total patients		57 *100*	9 *1 6*	15 *26*	33 *58*
Mean catalase activity . . .			21 %	34 %	34 %

[a]The parentheses indicate percentages based on fewer than 25 observations.

logically active and 64% deteriorated. The mean catalase activity for the patients who attained bacteriological quiescence was 21%, as compared with 34% for those who remained bacteriologically active and 34% for those who deteriorated. There is, thus, a slight suggestion that patients who had cultures with high catalase activity less frequently attained bacteriological quiescence than those who had cultures with lower catalase activity. However, the difference between the mean catalase activity of the cultures from the patients who attained quiescence and that of the cultures from those who remained bacteriologically active or who deteriorated does not attain significance ($P \sim 0.1$). The catalase activity of cultures was also estimated from the amount of gas liberated after immersion of a loopful of growth in a solution of hydrogen peroxide (qualitative method of Tuberculosis Chemotherapy Centre, 1959). The conclusions from the analysis of the results obtained by this method were very similar to those presented above for the semi-quantitative method.

The slight association between the attainment of bacteriologically quiescent disease and low catalase activity was also seen, though it did not attain significance, when the patients were stratified according to the extent of cavitation, but it was not apparent when they were stratified according to the degree of culture-positivity. This association was thus mainly a reflection of an association between catalase activity and the degree of culture-positivity at the end of the first year.

Isoniazid-resistant strains of tubercle bacilli which have high levels of resistance to isoniazid and low catalase activity are less virulent in the guinea-pig than strains with low levels of resistance and high catalase activity (Cohn et al., 1954; Mitchison, 1954; Freerksen & Meissner, 1956). It is therefore of particular interest to compare the response to treatment of patients who harboured these different types of isoniazid-resistant strains. Of the 16 patients who had cultures with both high or very high resistance to isoniazid according to the 20-colony MIC and a mean catalase activity of less than 20%, 19% had attained bacteriological quiescence by the end of the second year, 25% remained bacteriologically active and 56% had deteriorated to an extent that warranted a change of treatment. This is fairly similar to the response to treatment of the 14 patients who had cultures of moderate or low resistance and 20% or higher catalase activity, for 14% attained bacteriological quiescence, 43% remained bacteriologically active and 43% deteriorated.

Considering the equal-growth MIC, of the 13 patients who had cultures with high or very high

resistance and also less than 20% catalase activity, 23 % attained bacteriological quiescence, 15 % remained bacteriologically active and 62% deteriorated. This is not very different from the response of the 24 patients who had cultures with moderate or low resistance or " sensitive " organisms and 20 % or higher catalase activity, for 8 % attained bacteriological quiescence, 33 % remained bacteriologically active and 58% deteriorated. There is, thus, no evidence that cultures with very high isoniazid-resistance and little or no catalase activity were less virulent in patients than cultures with moderate or low resistance and greater catalase activity.

Susceptibility to hydrogen peroxide. Of the 57 patients, 35 had cultures obtained late in the first year which were tested for their susceptibility to hydrogen peroxide (Table 11). The cultures were isolated from 27 of these patients at 12 months, from one at 11 months and from seven at nine months. Of the 13 patients who had cultures of which less than 0.0001% of the tubercle bacilli survived exposure to hydrogen peroxide, 15% attained bacteriological quiescence, 15 % remained bacteriologically active and 69% deteriorated. The corresponding figures for the 22 patients who had cultures of which 0.0001% or more of the tubercle bacilli survived exposure were 5 %, 41% and 55 %, respectively. There was, thus, no association between response to treatment and the susceptibility of the cultures to hydrogen peroxide.

Virulence in the guinea-pig. Of the 57 patients, 52 had cultures obtained late in the first year which were tested for virulence in the guinea-pig, the cultures being isolated from 43 at 12 months, from two at 11 months and from seven at nine months. The response to treatment in the second year is related to the root-index of virulence in Table 12.

TABLE II

RESPONSE TO TREATMENT IN THE SECOND YEAR RELATED TO THE SUSCEPTIBILITY TO HYDROGEN PEROXIDE OF CULTURES ISOLATED AT THE END OF THE FIRST YEAR

Proportion of viable units which survived exposure to hydrogen peroxide (%)	Status of the disease at two years							
	Patients with cultures tested		Bacteriologically quiescent		Bacteriologically active		Radiographic deterioration warranting a change of treatment	
	No.	%	No.	%	No.	%	No.	%
Less than 0.0001	13	99	2	(15)[a]	2	(15)	9	(69)
0.0001-	7	100	0	(0)	2	(29)	5	(71)
0.01-	7	100	1	(14)	3	(43)	3	(43)
1.0 or more	8	100	0	(0)	4	(50)	4	(50)
Total patients . .	35	100	3	9	11	31	21	60

[a] The parentheses indicate percentages based on fewer than 25 observations.

TABLE 12

RESPONSE TO TREATMENT IN THE SECOND YEAR RELATED TO THE VIRULENCE IN THE GUINEA-PIG OF CULTURES ISOLATED AT THE END OF THE FIRST YEAR

Root-index of virulence of 9-, 11- or 12-month culture	Treatment series	Status of the disease at two years			
		Patients	Bacteriologically quiescent	Bacteriologically active	Radiographic deterioration warranting a change of treatment
		No. %	No. %	No. %	No. %
0.0-	PH	2	1	0	1
	HI-1	6	2	1	3
	HI-2	5	0	2	3
	H	19	2	2	15
	All series	32 101	5 1 6	5 16	22 6 9
	PH	2	0	1	1
	HI-1	6	0	2	4
	HI-2	5	1	3	1
	H	4	0	2	2
	All series	17 100	1 (6)[a]	8 (47)	8 (47)
0.9 or above	PH	0	0	0	0
	HI-1	0	0	0	0
	HI-2	0	0	0	0
	H	3	1	0	2
	All series	3 100	1 (33)	0 (0)	2 (67)
Total patients . . .		52 [b] 100	7 13	13 2 5	32 62
Mean root-index of virulence			0.47	0.62	0.53

[a] The parentheses indicate percentages based on fewer than 25 observations.

[b] Excluding five patients (1 PH, 1 HI-2, 3 H) who did not have virulence tests done on their 9-, 11- or 12-month culture.

Of the 32 patients who had cultures with a root-index of virulence of less than 0.60, 16 % had attained bacteriological quiescence by the end of the second year, 16% were bacteriologically active and 69% had deteriorated to an extent warranting a change of treatment. The corresponding proportions for the 17 patients who had cultures with a root-index of virulence between 0.60 and 0.89 were 6 %, 47 % and 47%. The mean root-index of virulence of the cultures from the seven patients who attained bacteriological quiescence was 0.47, from the 13 patients who remained bacteriologically active was 0.62 and from the 32 patients who had deteriorated was 0.53. Thus, there was little association between the response to treatment in the second year and the root-index of virulence of the cultures isolated at the end of the first year.

PROGRESS IN THE SECOND YEAR OF THE FOUR PATIENTS WHO HAD THEIR TREATMENT CHANGED AT THE END OF THE FIRST YEAR

Considering the three patients who, in error, did not continue on their initially prescribed chemotherapy, one (HI-2) received 200 mg of isoniazid daily and two (1 PH, 1 HI-2) received 500 mg of the placebo, calcium gluconate, daily. The HI-2 patient who received isoniazid yielded negative cultures throughout the second year, the last positive culture having been obtained at the eleventh month. At the end of the first year he had slight cavitation, a culture-positivity score of 0.1, and his cultures had very high resistance according to the 20-colony definition, moderate resistance according to the equal-growth definition, 0% catalase activity and a root-index of virulence of 0.57. The two patients who received the placebo had bacteriologically active disease at two years, but without evidence of deterioration. At the end of the first year the PH patient had moderate cavitation, a culture-positivity score of 0.3, moderate resistance according to both the 20-colony and equal-growth definitions, 30% catalase activity and a root-index of virulence of 0.36. The corresponding findings for the HI-2 patient were slight cavitation, a culture-positivity score of 0.2, moderate resistance according to both definitions, 35% catalase activity and a root-index of virulence of 0.54.

The H patient (see page 3) who had his treatment changed in the thirteenth month because of severe cor pulmonale, but who showed no evidence of deterioration of the tuberculosis, received streptomycin and PAS until his death from cardiac failure in the fifteenth month. His sputum remained culture-positive during the thirteenth and fourteenth months. At the end of the first year he had slight cavitation, a culture-positivity score of 2.3 and had cultures with very high resistance according to the 20-colony definition, moderate resistance according to the equal-growth definition, 13 % catalase activity and a root-index of virulence of 0.53.

OCCURRENCE OF DRUG-TOXICITY DURING THE SECOND YEAR

In the first year one (1%) of 90 PH, 13 (19 %) of 70 HI-1, six (9%) of 68 HI-2 and none of 87 H patients admitted to the study developed peripheral neuritis (Devadatta et al., 1960; Tuberculosis Chemotherapy Centre, 1960). Considering the 57 patients referred to in Table 3, two (one slow and one rapid inactivator of isoniazid) of the five PH patients had the prescribed chemotherapy for the whole of the second year, as did five (two slow and three rapid) of the 12 HI-1, seven (three slow and four rapid) of the 11 HI-2 and 10 (four slow and six rapid) of the 29 H patients. The durations of the prescribed chemotherapy in the remaining patients can be seen by reference to the last column of Table 3. There were no cases of peripheral neuritis during the second year among these 57 patients or in the four patients excluded from the main analysis, but one patient (HI-l), a slow inactivator, whose symptoms commenced in the eighth month of treatment was diagnosed by an independent assessor at the end of the sixteenth month of treatment as having peripheral neuritis.

DISCUSSION

An earlier report from the Centre (Tuberculosis Chemotherapy Centre, 1960) demonstrated that the combination of isoniazid plus PAS (the PH regimen) was substantially superior, in terms of the attainment of bacteriological quiescence, to any of the three regimens of isoniazid alone-namely, a moderate daily dosage (approximately 7.8-9.6 mg/kg) in one dose a day (the HI-l regimen); the same moderate daily

dosage in two doses a day (the HI-2 regimen); and a small daily dosage (approximately 3.9-5.5 mg/kg) in two doses a day (the H regimen). Thus, at one year 91% of 86 PH patients, as compared with 73 % of 64 HI-1, 58 % of 66 HI-2, and 44 % of 86 H patients, had bacteriologically quiescent disease or disease of doubtful bacteriological status which has since also been shown to be quiescent disease (Velu et al., 1961). The proportions of patients who died of tuberculosis or who had had their chemotherapy changed during the first year because of a clear-cut deterioration (radiographic in all except two cases) were 2% for the PH, 8 % for the HI-1, 23 % for the HI-2 and 21% for the H series. The HI-2 and H regimens were, therefore, clearly unsatisfactory in comparison with the PH regimen. The HI-1 series, on the other hand, although substantially inferior to the PH series in terms of the proportion of patients excreting tubercle bacilli (27%, as compared with 8%) showed a comparatively minor difference in terms of patients who had died or deteriorated and had their treatment changed (8 %, as compared with 2%). It is well established that isoniazid-resistant tubercle bacilli may have lowered virulence in the guinea-pig (Barnett, Bushby & Mitchison, 1953; Middlebrook & Cohn, 1953; Peizer, Widelock & Klein, 1953; Morse et al., 1954). A number of authorities consider that this applies in man also, and that the emergence of isoniazid-resistant strains during treatment, associated with persisting bacteriological positivity, is not necessarily a serious event (Deuschle et al., 1954; Middlebrook & Dressler, 1954; Oestreicher et al., 1955; McDermott, 1960). The relative merits of the PH and HI-1 regimens could therefore only be finally assessed by continuing the initially prescribed chemotherapy for the second year for the patients who had active disease at one year with resistant tubercle bacilli and establishing what course the disease followed. If the disease attained bacteriological quiescence, this would be evidence that bacteriological positivity with isoniazid-resistant organisms at one year was not a harmful state. Consequently the HI-1 regimen would approximate more closely to the PH regimen in efficacy. If however, the patients continued to remain bacteriologically positive and a substantial proportion had serious radiographic deterioration, then the danger of persisting bacteriological positivity would be established. Further information could be obtained on the general risk of persisting bacteriological positivity with isoniazid-resistant organisms by studying the HI-2 and H regimens and seeing whether the patients on them moved to bacteriologically quiescent disease or to radiographic deterioration in the second year.

In assessing the risk of bacteriological positivity with isoniazid-resistant organisms, it was necessary to take into account the observations of Oestreicher et al. (1955) that radiographic spreads which occur in patients undergoing treatment with isoniazid alone frequently resolve without a change of chemotherapy. A procedure of independent assessment was therefore followed, and the assessor reported a patient as needing a change of chemotherapy when he was satisfied that the radiographic progression of the disease was not of the type reported by Oestreicher et al. (1955). During the second year, three of six PH patients, seven of 12 HI-1, four of 13 HI-2 and 19 of 30 H patients–that is, 33 of 61 patients-had a radiographic deterioration warranting a change of treatment. (These figures include the three patients who received a low dosage of isoniazid or a placebo in the second year and did not deteriorate, and the patient who died of cor pulmonale.) The amalgamated findings for tuberculous deaths and deteriorations in the 2-year period in the patients whilst on their initially prescribed chemotherapy consequently increased to five (6 %) of 86 PH, 12 (19 %) of 64 HI-1, 19 (29 %) of 66 HI-2 and 37 (43 %) of 86 H patients.

Only nine of the 57 patients who continued on their originally prescribed chemotherapy in the second year attained bacteriologically quiescent disease; six had yielded the last positive culture towards the end of the first year, the remaining three doing so at the twelfth or thirteenth month. Not one patient who was bacteriologically positive at fourteen months subsequently attained bacteriological quiescence. Of the 57 patients, no less than 33 had a radiographic deterioration warranting a change of chemotherapy. Thus, a clear picture emerged–namely, that bacteriological positivity with isoniazid-resistant organisms at one year, irrespective of their level of resistance to isoniazid or their catalase activity, was in nearly all instances followed by deterioration in the second year or persisting positivity at two years. It is therefore reasonable to assume that the persisting positivity at two years would also eventually lead to deterioration.

The only assessments at one year which were related to the therapeutic response in the second year were the extent of cavitation and the degree of culture-positivity. Of the nine patients who attained bacteriological quiescence, eight had very low

culture-positivity scores (six having just started the period of bacteriological quiescence towards the end of the first year) and six had no cavitation at the end of the first year. However, it was not possible to forecast with any degree of certainty which of the patients who remained bacteriologically positive after one year's treatment would attain bacteriological quiescence during the second year of treatment, for only six of the 12 patients who had both very low positivity scores and no cavitation attained bacteriological quiescence.

Detailed analyses did not demonstrate any clear evidence of an association between the response to treatment in the second year and the level of isoniazid-resistance, the catalase activity, the susceptibility to hydrogen peroxide or the degree of virulence in the guinea-pig of cultures isolated at the end of the first year. It will be appreciated that the population of patients studied in this report is a selected one and that a number of patients who died or who deteriorated radiographically and had their treatment changed in the first year do not appear in the analysis; nor do patients who yielded isoniazid-resistant strains in the first year and then attained bacteriological quiescence before the end of the year. The conclusions as to the influence of virulence, the degree of isoniazid-resistance, the catalase activity and hydrogen peroxide susceptibility may not necessarily apply to the full population of patients admitted to the study or to the influence of these factors in the first year. A further report will be published on the characteristics of the isoniazid-resistant organisms isolated during the first year and their influence on the response of the patients.

It may be concluded that persisting bacteriological positivity at the end of one year's treatment, even if the patient is excreting highly isoniazid-resistant, catalase-negative tubercle bacilli, is likely to lead to serious radiographic deterioration. This finding is in clear contradistinction to those of Oestreicher et al. (1955). The finding that isoniazid-resistant infections are not self-limiting has considerable implications for mass therapy in developing countries. If such therapy is with isoniazid alone it is very likely that many cases with drug-resistant infections will be produced and that the disease in these patients will follow a deteriorating course. This observation underlines the importance of the use, whenever possible, of effective combined chemotherapy and of the need for finding effective, inexpensive and acceptable alternative regimens for the treatment of pulmonary tuberculosis.

SUMMARY

1. Of the 315 patients in the main analysis of a controlled comparison of isoniazid plus PAS with three regimens of isoniazid alone, 61 had bacteriologically active or relapsed disease with isoniazid-resistant organisms at the end of the first year of treatment. This paper has presented the results of a study of the response to treatment in the second year of these 61 patients, 57 of whom continued on their originally allocated treatment (five on isoniazid 4.5 mg/kg plus PAS 0.23 g/kg daily, divided into two doses; 12 on isoniazid 8.7 mg/kg daily, in one dose; 11 on isoniazid 8.7 mg/kg daily, divided into two doses; and 29 on isoniazid 4.5 mg/kg daily, divided into two doses). The study was undertaken in order to determine the prognosis of persisting bacteriological positivity at one year with isoniazid-resistant tubercle bacilli.

2. Of the 57 patients, nine (16 %) attained bacteriological quiescence, 15 (26%) had bacteriologically active disease at the end of the second year and 33 (58 %) had had a serious radiographic deterioration which, in the opinion of an independent assessor, warranted a change of treatment.

3. The response to treatment in the second year was similar in each of the four treatment series.

4. There was an association between the response to treatment in the second year and both the extent of cavitation and the degree of culture-positivity at the end of the first year. Patients with no cavitation or whose specimens were weakly positive on culture more frequently attained bacteriological quiescence and, conversely, patients with cavitation or with heavily positive cultures more frequently deteriorated radiographically. However, six of the nine patients who achieved bacteriological quiescence had already commenced the period of persisting bacteriological negativity in the first year; the other three did so before 14 months.

5. There was no clear association between response to treatment in the second year and the number of lung zones involved in disease, the total radiographic extent of the lesion, the level of resist-

ance to isoniazid according to two definitions, the catalase activity, the susceptibility to hydrogen peroxide or the virulence in the guinea-pig of cultures isolated in the last months of the first year.

6. It is concluded that the disease in patients with persisting bacteriological positivity with isoniazid-resistant organisms at the end of one year of treatment with isoniazid, alone or in combination with PAS, is likely to deteriorate irrespective of the level of resistance to isoniazid or of the catalase activity of the cultures of tubercle bacilli. The object of treatment of patients with bacteriologically positive pulmonary tuberculosis must therefore be to render the sputum culture-negative in all instances.

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RÉSUMÉ

A la fin de la première année de traitement contre la tuberculose, 61 des 315 malades considérés dans l'analyse générale d'une étude comparée de régimes thérapeutiques (isoniazide+PAS, et trois régimes d'isoniazide seul) étaient bacillaires, qu'ils le soient demeurés ou aient été victime d'une rechute. Leurs bacilles étaient résistants à l'isoniazide.

Dans cet article, les auteurs examinent la réponse au traitement de ces 61 malades, au cours de la deuxième année de traitement. Cinquante-sept d'entre eux continuèrent le traitement qui leur avait été assigné (5 recevant 4,5 mg/kg/jour d'isoniazide + 0,23 g/jour de PAS, administré en deux doses; 12 recevant 8,7 mg/kg/jour en une dose; 11 recevant 8,7 mg/kg/jour en deux doses; 29 recevant 4,5 mg/kg/jour en deux doses). L'intention des auteurs était de préciser le pronostic des malades qui, après une année de traitement, présentaient des bacilles résistants à l'isoniazide.

Sur ces 57 malades, 9 (16%) avaient atteint la quiescence bactériologique à la fin de la deuxième année; 15 (26%) étaient encore bacillaires et chez 33 (58%), la situation indiquée par l'examen radiologique s'aggravait de telle sorte qu'un examinateur indépendant jugea nécessaire de changer le traitement de ces malades.

La réponse au traitement, la deuxième année, était analogue au sein des quatre séries de traitement.

On nota une relation entre la réponse au traitement la deuxième année, l'étendue des lésions cavitaires et le degré de positivité des cultures à la fin de la première année. Les malades non cavitaires ou dont les crachats n'étaient que faiblement positifs à la culture atteignaient plus fréquemment l'état de quiescence bacillaire, tandis que les malades à lésions cavitaires étendues et à cultures fortement positives présentaient plus souvent une détérioration que révélait l'examen radiologique. Tous les malades qui atteignirent le stade de quiescence bactériologique avaient déjà présenté une négativité à la culture dès la première année, sauf un pour qui cette négativité ne se manifesta qu'après treize mois.

Il n'y eut pas de relation évidente entre la réponse au traitement la seconde année et le nombre des zones pulmonaires intéressées, l'étendue des lésions (à l'examen radiologique), le niveau de résistance à l'isoniazide (selon deux définitions), l'activité de la catalase, la sensibilité à l'eau oxygénée ou la virulence pour le cobaye des cultures isolées durant les derniers mois de la première année.

En conclusion, les auteurs admettent que les malades bacillaires de façon permanente — présentant des bacilles résistants à l'isoniazide — à la fin d'une année de traitement par l'isoniazide+PAS ou l'isoniazide seul, courent le risque de voir leur maladie s'aggraver, quel que soit le niveau de résistance de leurs bacilles ou l'activité de la catalase des cultures. Le traitement des malades atteints de tuberculose pulmonaire et bactériologiquement positifs doit tendre, dans tous les cas, à rendre les crachats bactériologiquement négatifs.

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