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Resistant and Sensitive Strains of *Mycobacterium tuberculosis* Found in Repeated Surveys among a South Indian Rural Population *

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The findings in a highly selected group of patients, such as those attending clinics or sanatoria, cannot be used as the basis for assessing the true prevalence of strains of Mycobacterium tuberculosis *with acquired or primary resistance or of sensitive strains in a community. The present report describes the prevalence of such strains as found in 3 successive surveys in a sizeable random sample of villages in a South Indian district. Changes in the status of cases with such strains from an earlier survey to a later one and the status at an earlier round of cases found at a later one are also described.*

The prevalence of tuberculous infection among household contacts of cases with acquired resistance to isoniazid was significantly higher than that among contacts of cases with primary resistance or of those with sensitive cultures. This is probably due to the longer duration of sputum positivity of the former at the time of diagnosis. But infectivity, as judged by the incidence of new infections among household contacts, was generally less for cases with acquired or primary resistance than for cases with sensitive cultures, though the difference was not statistically significant.

A large number of culture-positive cases, especially those with primary resistance, had no radiological evidence of active pulmonary tuberculosis. The prevalence of primary resistance was very high among certain categories of cases, and the differences between cases with primary resistance and those with acquired resistance were many and large. It is suggested that this could be due to some of the primary resistant cultures being those of atypical mycobacteria, despite positivity in the niacin test.

There was a significant increase in the number of cases with acquired resistance to isoniazid at the third survey round owing to irregular treatment with that drug after the second round. The prevalence of primary resistance at the 3 rounds was almost the same.

INTRODUCTION

The degree of the risk of infection and disease in man from drug-resistant strains of *Mycobacterium tuberculosis* is not clear. An increase in the prevalence of strains with primary resistance could indicate the extent of such a risk, while an increase in the prevalence of acquired resistance may reflect only limitations of the treatment available. If primary resistance does not increase over a long period, an increase in secondary or acquired resistance could be considered to be a problem of the individual patient only, involving little or no danger to the community. It is therefore of considerable importance to keep a continuous watch for any increase in primary resistance. Owing to the chronic nature of the disease and the low incidence of infection, such an increase may take a long time to manifest itself. But until

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such a time as the public health risk of resistant strains can be completely ruled out, it would be prudent to adopt a cautious attitude towards any increase in resistant strains, even though such strains are less pathogenic than sensitive strains to guinea-pigs (Middlebrook & Dressler, 1954) and possibly to man (Šerman, 1963).

An increase in the prevalence in the community of resistant strains, either primary or acquired, cannot be accurately estimated from data derived from the highly selected and possibly variable population represented by patients reporting to clinics or sanatoria. The present study is based on 3 consecutive surveys in a random sample of villages in Bangalore District, South India, and gives the prevalence of primary and acquired resistance and of sensitive strains at each survey, changes in the status of cases with such strains from one survey to another and their infectivity among household contacts. Many anomalous findings are reported, on the basis of which a suggestion is made that a proportion of the cultures with primary resistance probably represented atypical mycobacteria, in spite of their being niacin-positive.

METHODS AND MATERIAL

A random sample of 133 villages from among the 734 villages in 3 taluks of Bangalore District, South India, was surveyed from May 1961 to July 1963 (Round I) by the National Tuberculosis Institute, Bangalore. Of these villages, 119 were resurveyed (Round II) after an average interval of 18 months, and 14 (population about 10 000) after an average interval of 9½ months, the aim in these 14 villages being to study changes after a shorter interval. A third survey (Round III) of *all the 133 villages* was carried out after an average interval of 18 months from the second survey. No tuberculin testing or BCG vaccination had previously been carried out in these villages. Survey techniques have been described in detail earlier (Raj Narain et al., 1963). Briefly, the procedures were as follows.

All persons were registered and offered a tuberculin test with 1 TU of RT 23 in 0.1 ml phosphate buffer containing 0.005% Tween 80. In each survey tuberculin testing was done at a different site in the forearm, the longitudinal diameter of the reaction being recorded in millimetres 3–4 days after the testing. Those 5 years of age or more were offered, in addition, a 70-mm photofluorogram. For all photofluorograms, readings were recorded independently by two readers in one of the following 5 categories:

O. *No abnormality.*

A. *Probably non-tuberculous:* All lesions considered of non-tuberculous origin.

B. *Probably tuberculous but inactive:* All scars and other healed lesions except calcifications (some doubtful shadows, if they are recorded at all, may be classified as inactive).

C. *Probably tuberculous, possibly active:* Lesion appearing to be of tuberculous nature but without a definite cavity and not extensive.

D. *Probably tuberculous and active:* The lesion appears to be of tuberculous nature, may be extensive, may be bilateral, or a definite cavity is present.

Cases assigned to either of the last two categories, namely, C or D, by *both* readers are referred to as "X-ray cases". When the two readers differed, a third reader's reading was taken as final; the third reader was called upon only where there was a difference between the first two. Cases confirmed as C or D by him were also considered as X-ray cases.

All those with any kind of abnormality recorded by either of the two readers, even those with only suspected pathology (but not those with calcification only) and those with unsatisfactory photofluorograms or those who could not be X-rayed owing to physical disability were eligible for collection of 2 specimens of sputum; one "spot" in the evening and another "overnight" in the morning. All those eligible for sputum examination in an earlier survey were also eligible for sputum examination in subsequent surveys. Each specimen of sputum was examined independently by fluorescence microscopy, by the Ziehl-Neelsen technique, and by culture on 2 slopes of Löwenstein-Jensen medium without potato starch, after homogenization with 4% sodium hydroxide.

For specimens showing growth on culture at 4 weeks, a colony count, based on the culture tube showing the largest number, was made; the exact number was recorded if the count was less than 20. A count of 20 colonies or more is described as

TABLE 1
COVERAGES AT EACH ROUND FOR X-RAY AND SPUTUM EXAMINATION IN 133 VILLAGES

Round	Registered *de jure* population [a]	No. excluded owing to presence of BCG scar	X-ray examination			Sputum examination		
			No. eligible	X-rayed		No. eligible	Examined	
				No.	%		No.	%
I	73 689	2 328	60 389	52 215	86.5	7 118	6 453	90.7
II	74 234	2 803	59 832	49 273	82.4	12 725	10 894	85.6
III	74 978	2 738	60 911	49 723	81.6	18 278	15 760	86.2

[a] Figures include those with BCG scar.

positive. For cultures that showed no growth or only faint growth at 4 weeks, a colony count was made at 8 weeks.

All positive cultures were identified by subculturing and observing growth at room temperature, production of pigment in the dark and after exposure to daylight, rate of growth at 37°C, peroxidase and catalase reactions and niacin production. The last test was introduced about 7 months after the start of the first survey; the results of this test are therefore not available for 64 cases from Round I, including 8 with resistant cultures. Only cultures identified as being of tubercle bacilli of the human type have been included in this report.

Sensitivity tests against several concentrations of isoniazid, streptomycin and *p*-aminosalicylic acid (PAS) were also carried out (for details, see Rao et al., 1966). A minimal inhibitory concentration (MIC) of 1 μg/ml restricting growth on culture to less than 20 colonies or a resistance ratio (RR) of at least 4 for streptomycin or PAS was the criterion for describing cultures as resistant. The degree of resistance is indicated wherever relevant. If a culture resistant to any drug was grown from only 1 of the 2 sputum specimens, even though the other specimen yielded a sensitive culture, the case is reported as being one with resistant culture.

History of previous treatment was unfortunately not recorded during the first survey. A history of treatment was collected at the time of the second and the third surveys by the tuberculin test reader for all bacillary cases diagnosed during an earlier survey and for those read as X-ray category C or D by either reader during either of the later surveys. Some 2-3 months after the second survey, trained sociological workers collected histories in great detail from bacillary cases and controls. All available sputum-positive cases from all 3 rounds were later interviewed by a medical officer. Thus for most of the patients a history is available at 4 different points of time as collected by 4 different persons.

Owing to lack of resources, treatment could not be offered to patients at the end of Round I. Not many facilities for treatment are available in the area. During the second and third surveys all sputum-positive cases from the previous round, if available, and all cases of X-ray category D as recorded by both readers were given isoniazid tablets in sufficient quantity to last one month. Arrangements were made for a further free supply of tablets from the nearest medical facility.

A resistant culture in a case with a history of treatment of at least 15 days with the relevant drug or drugs at any time prior to sputum examination is reported here as being due to acquired resistance.

STUDY POPULATION

All permanent residents, including those temporarily away at the time of registration, constitute the study population at each round. The coverages at the 3 rounds for X-ray and sputum examinations are shown in Table 1. The numbers excluded at each round owing to the presence of a BCG scar are also shown. Among those excluded at Round I were 5 cases with sensitive cultures; at Round II, 2 cases with resistant and 4 with sensitive cultures; at Round III, 2 cases with resistant and 3 with sensitive cultures. One of the excluded cases was common to all 3 rounds; the culture was sensitive at Rounds I and II and resistant to isoniazid at Round III. Two cases were common to Rounds II and III; in one, the culture was resistant at Round II but both had sensitive cultures at Round III.

TABLE 2
CULTURE-POSITIVE CASES AND CASES WITH RESISTANT CULTURES AT EACH OF THE 3 ROUNDS

Round	Total culture-positive cases	Cases with resistant cultures	Cases with culture resistant to					
			Isoniazid alone	Isoniazid and PAS	Isoniazid and streptomycin	All 3 drugs	Streptomycin alone [a]	PAS alone [a]
			For 119 villages [b]					
Round I	179 [c]	25	10	5	2	3	3	2
Round II	167	32	17	6	4	—	—	5
Round III	156	48	31	5	4	—	5	3
			For 14 villages [b]					
Round I	20	5	3	—	—	1	—	1
Round II	27	4	1	1	1	—	—	1
Round III	21	5	4	1	—	—	—	—

[a] No culture was found resistant to both streptomycin and PAS.
[b] See text.
[c] Excludes 1 case for which sensitivity test was not done.

FINDINGS

PREVALENCE AT EACH ROUND

Table 2 shows the number of culture-positive cases and of cases with resistant cultures at each of the 3 rounds separately for the 2 sets of villages. The total culture-positive cases, in both sets of villages, as judged by the results with either one or both sputum specimens, numbered 199, 194 and 177 respectively at the 3 rounds, constituting prevalence rates of 3.8, 4.0 and 3.7 per thousand of those who were X-rayed, whose sputa were examined or both. The total cases with cultures resistant to one or more drugs numbered 30, 36 and 53 respectively, i.e., 15.1%, 18.6% and 30% of the culture-positives at the 3 rounds respectively. These included 13, 18 and 35 resistant to isoniazid alone. The marked increase in the number of patients with resistant cultures at Round III is due to an increase in resistance to isoniazid only. The numbers of persons with strains resistant to PAS or streptomycin were 6, 6 and 8 respectively in the 3 rounds.

A comparison of the cases with primary or acquired resistance and of those with sensitive cultures showed no significant difference in the sex ratio, in the number of tuberculin-negatives (either as a whole or by sex) or in the mean size of induration to the tuberculin test. These findings have not been tabulated.

RESULTS OF SENSITIVITY TESTS FOR THE 2 SPUTUM SPECIMENS

From each person 2 specimens of sputum were collected. Table 3 shows the cases with cultures sensitive to or having primary or acquired resistance to any of the 3 drugs; the results are shown for the 2 specimens together and separately. Cultures resistant to more than one drug have been included in the assessment for each drug and the results of all 3 rounds have been pooled.

The same sensitivity result with both specimens was more often encountered among cultures with acquired resistance to isoniazid or streptomycin than among those with primary resistance or which were sensitive; the difference for those with resistance to isoniazid + was highly significant ($P < 0.01$) and for those with resistance to streptomycin + was significant ($P < 0.05$).[1] Cultures with primary resis-

[1] Throughout this paper " resistance to isoniazid +" is used to mean resistance to isoniazid alone or in combination with either or both of the other two drugs, " streptomycin +" to indicate resistance to streptomycin alone or in combination, and " PAS +" to indicate resistance to PAS alone or in combination.

TABLE 3

RESULTS OF SENSITIVITY TESTS TO DIFFERENT DRUGS BY 2 SPUTUM SPECIMENS, POOLED FOR ALL 3 ROUNDS IN 133 VILLAGES

Drug to which resistant [a]	Primary or acquired resistance		No. of cases with cultures resistant or sensitive by:			
			Either specimen of sputum	Both specimens of sputum	1 specimen and negative by the other	1 specimen and sensitive by the other (resistant cultures only)
Isoniazid+	Primary	No.[b]	22	2	13	7
		%	100	9.1	59.1	31.8
	Acquired	No.[c]	72	50	17	5
		%	100	69.4	23.6	6.9
Streptomycin+	Primary	No.	11	4	5	2
		%	100	36.4	45.4	18.2
	Acquired	No.	12	10	—	2
		%	100	83.3	—	16.7
PAS+	Primary	No.	21	2	13	6
		%	100	9.5	61.9	28.6
	Acquired	No.[d]	11	3	—	8
		%	100	27.3	—	72.7
Sensitive to all 3 drugs		No.	315	141	174	—
		%	100	44.8	55.2	—

[a] The sign + after the name of the drug indicates resistance to that drug either alone or in combination with either of the other 2 drugs.

[b] Excludes 1 case (at Round I), for which sensitivity test was not done for the overnight sample; the spot sample was resistant with a MIC of 1 μg/ml.

[c] Excludes 4 cases for which history of treatment was not available.

[d] Excludes 2 cases for which history of treatment was not available.

tance were more often negative or sensitive by the second specimen than cultures with acquired resistance. Except for streptomycin + in the last column of Table 3, all differences from the cultures with acquired resistance were significant, mostly at the 99% level of confidence.

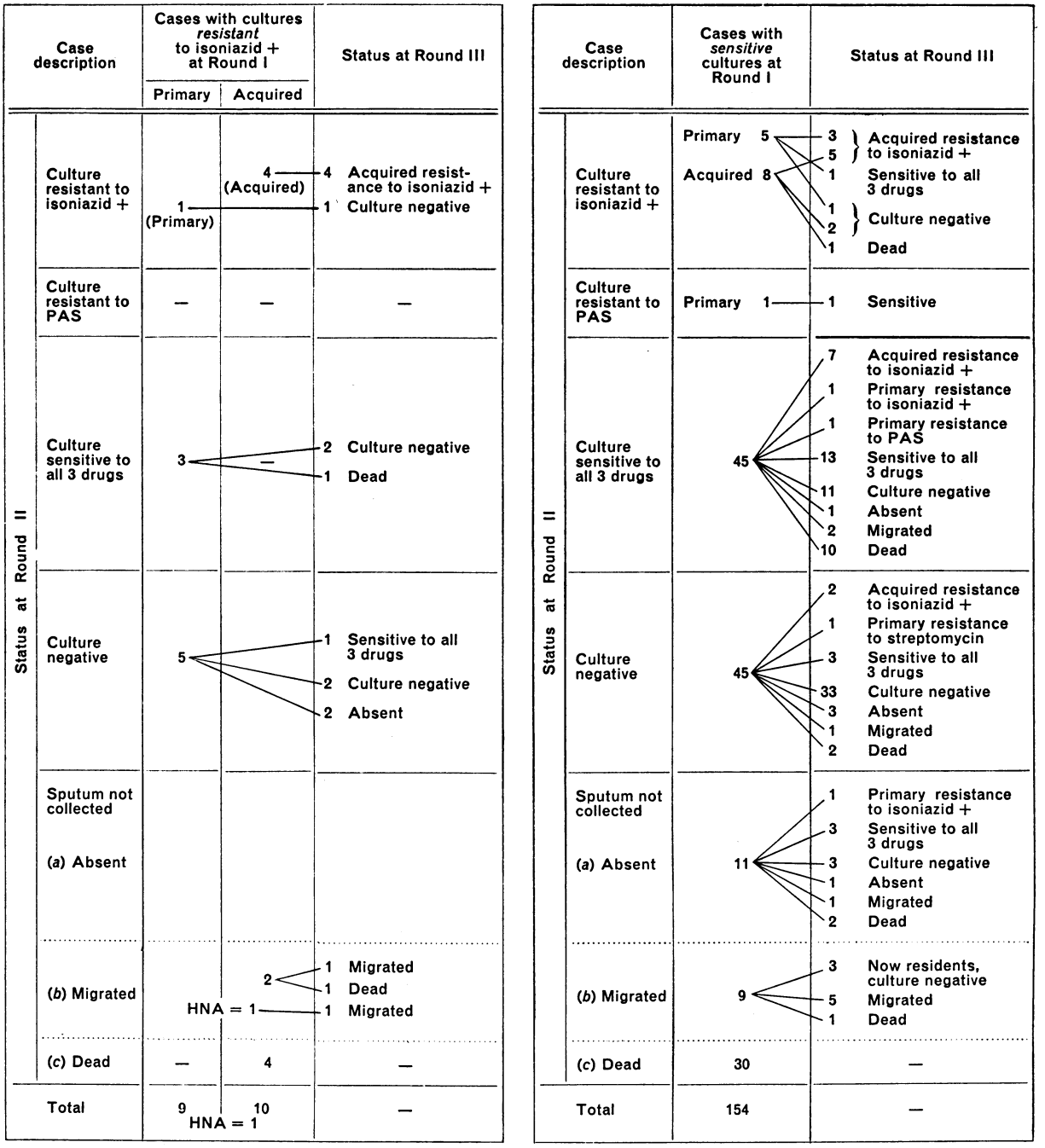
The percentage of cultures with acquired resistance found in *both* specimens was less for PAS+ than for either isoniazid+ or streptomycin+. If consistency of findings in the 2 specimens is any indication of the reliability of the sensitivity tests, then the finding of cultures resistant to PAS+ would appear to be a considerably less reliable criterion of resistance than finding cultures resistant to the other drugs.

CHANGES WITH TIME IN CASES WITH RESISTANT AND SENSITIVE CULTURES

The status of cases at a subsequent round shows the mortality or other naturally occurring changes with time, while the status at an earlier round of cases found at a later one shows the changes from the reverse direction, especially if there were any differences in the sputum-positivity rate at the earlier round or rounds in different types of cases. To keep the interval between rounds uniform, only 119 villages are considered in Fig. 1, 2, 3 and 4. Cases resistant to PAS or streptomycin, being too small in number, have not been included, but are described separately.

FIG. 1

STATUS OF ROUND-I CASES AT ROUNDS II AND III, SEPARATELY FOR THOSE WITH PRIMARY OR ACQUIRED RESISTANCE TO ISONIAZID+ AND FOR THOSE WITH SENSITIVE CULTURES, IN 119 VILLAGES[a]



[a] Radiating lines indicate status at Round III. "Isoniazid +" refers to isoniazid either alone or in combination with either of the other 2 drugs (streptomycin and PAS). HNA = History of treatment not available.

Status at a subsequent round

Fig. 1 shows the status of Round-I cases at Rounds II and III and Fig. 2 the status at Round III of the new cases of Round II.

Status of Round-I cases at Rounds II and III. Of the 9 cases at Round I with primary resistance to isoniazid alone or in combination with streptomycin and PAS (i.e., isoniazid+), 5 had negative cultures at Round II, 3 had sensitive cultures and 1 had primary resistance again (Fig. 1). None had died. At Round III, of the 5 with negative cultures at Round II, 1 yielded a sensitive culture, 2 had negative cultures and 2 could not be examined. Of the 3 with sensitive cultures at Round II, 2 were negative and 1 had died. For the case with primary resistance at Rounds I and II, the culture was negative at Round III.

Of the 10 cases with acquired resistance at Round I, 4 had died by Round II, 2 had migrated and 4 continued to yield resistant cultures. These 4 were also resistant at Round III (i.e., acquired resistance at all 3 rounds).

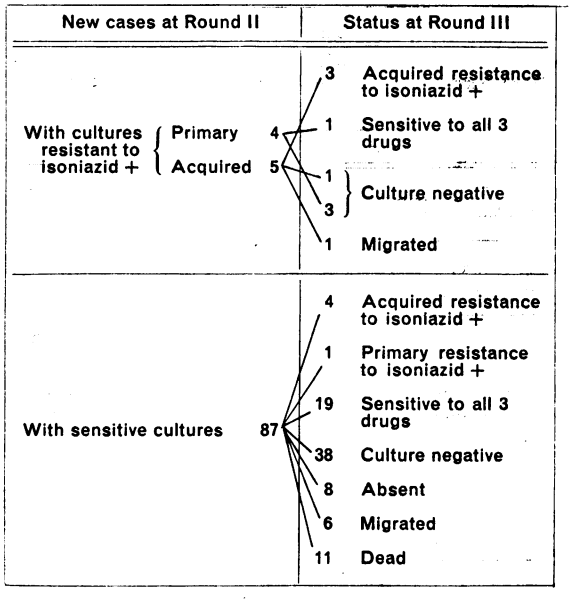
Fig. 1 also shows the status at Rounds II and III of 154 cases with sensitive cultures at Round I. At Round II, 30 had died, 8 yielded cultures with acquired resistance and 6 (5+1) yielded cultures with primary resistance; 45 had sensitive cultures, and another 45 negative cultures; 20 could not be examined. The status of these categories at Round III is also shown. Of the 8 with acquired resistance, 5 continued to show resistance, 2 were culture-negative and 1 had died. Of the 6 (5+1) with primary resistance, 3 had acquired resistance at Round III, 1 was culture-negative and 2 yielded sensitive cultures. Of the 45 with sensitive cultures, 7 had acquired resistance and 2 primary resistance at Round III, while 13 had sensitive cultures; 11 were culture-negative and 10 had died.

Of the 45 culture-negatives at Round II, 6 were culture-positive at Round III—2 with acquired and 1 with primary resistance and 3 with sensitive cultures. Of the 11 absent from examination at Round II, 4 were culture-positive at Round III; this included 1 with primary resistance to isoniazid+.

Status of Round-II cases at Round III. There were 162 culture-positive cases in 119 villages at Round II; 96 were culture-positive for the first time and are shown in Fig. 2. Four had primary and 5 acquired resistance, while 87 had sensitive cultures. Of the 4 with primary resistance, 3 were culture-negative at Round III and 1 yielded a sensitive culture. Of the 5 with acquired resistance, 3 continued to show such resistance. Of the 87 with sensitive cultures, 4 had acquired and 1 had primary resistance at Round III; 19 yielded sensitive cultures, 38 were culture-negative and 11 had died.

FIG. 2

STATUS OF NEW CULTURE-POSITIVE CASES OF ROUND II AT ROUND III

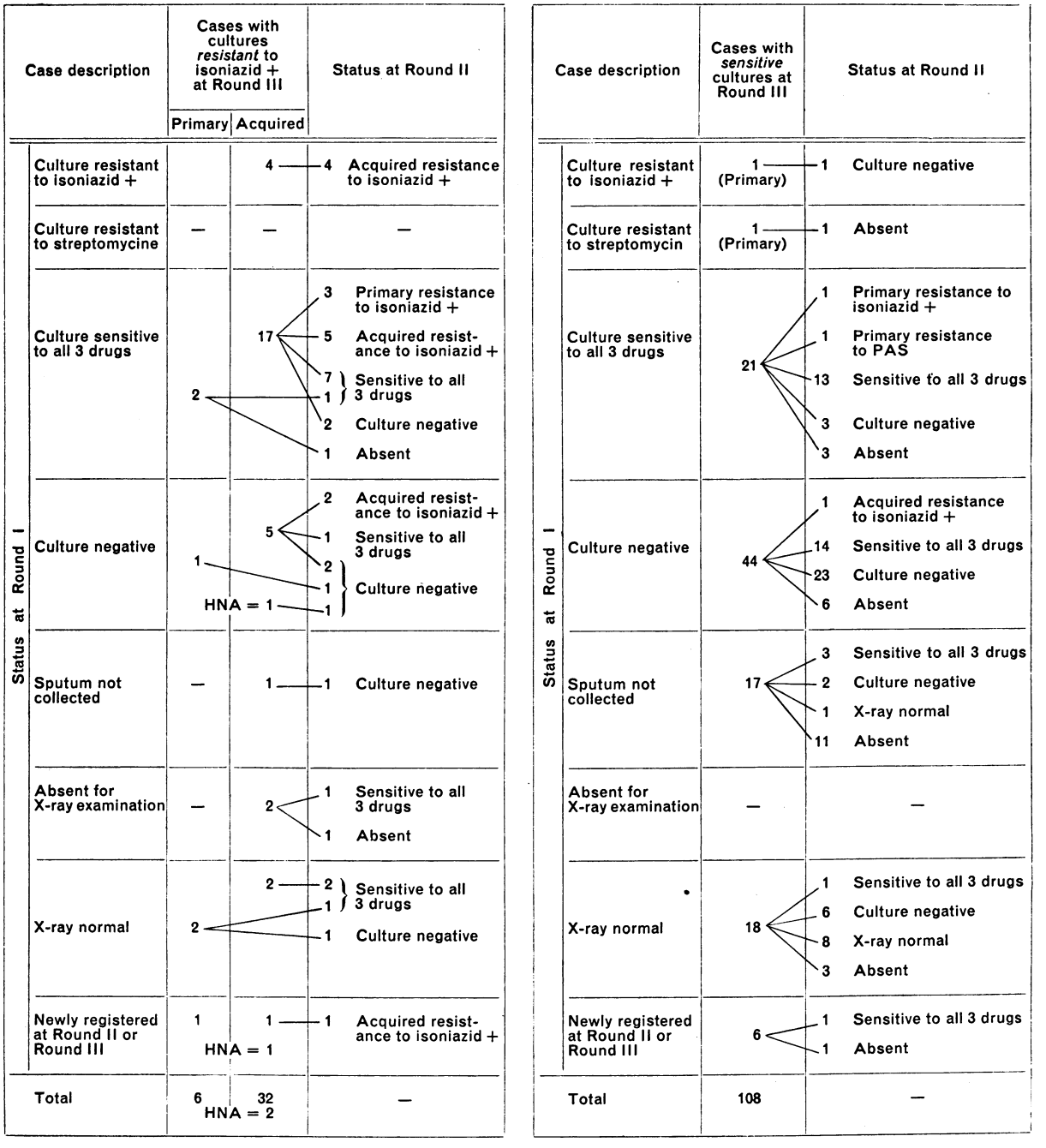


Mortality as against sensitivity status. From Fig. 1 it will be seen that cases with acquired resistance at Round I had the highest mortality. The difference between them and cases with primary resistance was significant ($P<0.05$) for deaths at both Round II and Round III, but the difference was not significant between them and those with sensitive cultures nor between those with primary resistance and those with sensitive cultures.

Conversion to negative cultures. At a subsequent round, cases with primary resistance became culture-negative most often and cases with acquired resistance least often; the difference between the two was significant from Round I to both Round II and Round III ($P<0.05$). The difference between cases with sensitive cultures and those with acquired resistance was significant only for results from Round I to Round III ($P<0.05$); the difference between those with primary resistance and those with sensitive cultures was not significant.

FIG. 3

STATUS OF ROUND-III CASES AT ROUNDS I AND II, SEPARATELY FOR THOSE WITH PRIMARY OR ACQUIRED RESISTANCE TO ISONIAZID+ AND FOR THOSE WITH SENSITIVE CULTURES, IN 119 VILLAGES [a]



[a] Radiating lines indicate status at Round II. "Isoniazid +" refers to isoniazid either alone or in combination with either of the other 2 drugs (streptomycin and PAS). HNA = History of treatment not available.

Cases with cultures resistant to streptomycin or PAS. These are not shown in Fig. 1 and Fig. 2. Of 5 such cases at Round I (Table 2), 2 had acquired and 3 had primary resistance. Four had died by Round II; radiologically all 4 were advanced cases of pulmonary tuberculosis. The fifth, with primary resistance, could not be examined at Round II, but yielded sensitive cultures at Round III.

All the 5 cases at Round II (Table 2) had primary resistance to PAS. At Round III 2 were culture-negative, 1 yielded a sensitive culture, 1 had died and 1 had migrated.

Status at an earlier round

Fig. 3 shows the status of Round-III cases at Rounds II and I and Fig. 4 that of Round-II cases at Round I.

Status of Round-III cases at Rounds II and I. Of the 6 cases with primary resistance (Fig. 3), 2 had sensitive cultures at Round I and 2 at Round II, 1 being common to both rounds.

Of the 32 cases with acquired resistance at Round III, 4 had acquired resistance at both Rounds I and II; in addition 3 had primary and 8 had acquired resistance at Round II; 7 were sensitive at both Rounds I and II, 4 were sensitive at Round II only and 2 were sensitive at Round I only. Thus of the 32 cases with acquired resistance at Round III, 28, or 88%, had positive cultures at Round I or Round II.

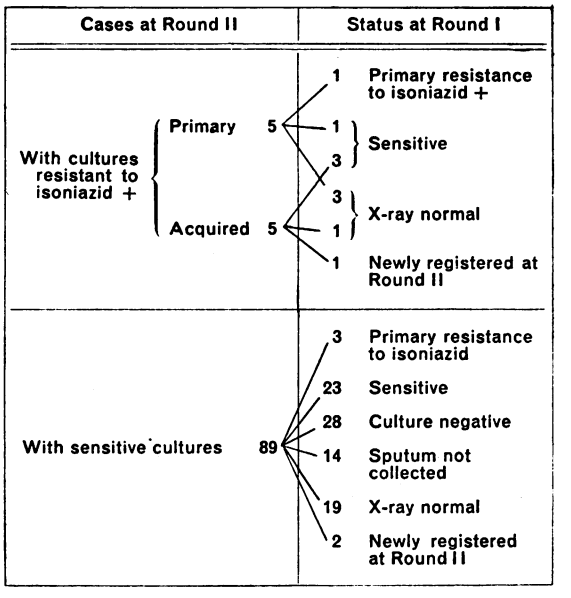
Of the 108 sensitive cultures at Round III, 21 had sensitive and 2 had primary resistant cultures at Round I. Twenty more, including 1 with acquired resistance, were culture-positive at Round II: thus 43, or 40%, of the 108 were culture-positive at Round I or Round II.

Status of Round-II cases at Round I. The status at Round I of those Round-II cases which have not been included in Fig. 3 is shown in Fig. 4. Of the 5 cases at Round II with primary resistance to isoniazid+, 1 had a sensitive culture at Round I, 1 had a culture with primary resistance and 3 were X-ray normal (not eligible for sputum collection) at Round I. Of the 5 with acquired resistance at Round II, 3 had sensitive cultures at Round I. Of the 89 cases with sensitive cultures at Round II, 26 only (3 with primary resistance and 23 with sensitive cultures) were culture-positive at Round I.

Thus, as seen in Fig. 3 and 4, at the time of diagnosis, a much larger proportion of the cases diagnosed at one of the later rounds as having acquired

FIG. 4

STATUS AT ROUND I OF ROUND-II CASES WHICH HAVE NOT BEEN INCLUDED IN FIG. 3



resistance than as having sensitive cultures had at an earlier round been sputum-positive, either with resistant or with sensitive cultures. The difference is significant ($P<0.01$) for Round-III cases at both Round II and Round I and for Round-II cases at Round I; that is to say, cases with acquired resistance had been culture-positive longer than other cases. Cases with primary resistance occupied a somewhat intermediate position in this regard, although the difference between them and those with sensitive cultures was not statistically significant for the 2 intervals.

Cases with cultures resistant to streptomycin or PAS. Of the 8 who were resistant to streptomycin or PAS at Round III (Table 2), 7 had primary resistance and in 1 history was not available. Of the 7, 2 had sensitive cultures at Round I (and 1 at Round II also), 3 had normal X-rays at both Round I and Round II and thus were not eligible for sputum examination, and 2 were newly registered at Round III. The case for which a history was not available had a doubtful shadow at Round I, assessed by one reader only; sputum could not be collected at Rounds I or II.

All the 5 cases at Round II had primary resistance to PAS. At Round I, 2 were culture-negative, 1 had

TABLE 4

CASES WITH PRIMARY OR ACQUIRED RESISTANCE OR WITH SENSITIVE CULTURES AT EACH OF THE 3 ROUNDS IN 133 VILLAGES

Round	Total cultures positive		Primary resistance			Acquired resistance			Sensitive to all 3 drugs
			To any of the 3 drugs	To iso-niazid+	To PAS or strepto-mycin	To any of the 3 drugs	To iso-niazid+	To PAS or strepto-mycin	
I	199	No.	14	10 [a]	4	14	12	2	169
		%	7.0	5.0	2.0	7.0	6.0	1.0	84.9
II	194	No.	16	10	6	20	20	—	158
		%	8.2	5.1	3.1	10.3	10.3	—	81.4
III	177	No.	13	6 [a]	7 [b]	37	37	—	124
		%	7.3	3.4	4.0	20.9	20.9	—	70.1

[a] Excludes 2 cases with cultures resistant to isoniazid+ for whom history of treatment was not available.
[b] Excludes 1 case with cultures resistant to PAS for whom history of treatment was not available.

a sensitive culture, 1 was X-ray normal and the fifth could not be examined.

It may be seen that a vast majority of the cultures resistant to streptomycin or PAS at all 3 rounds had primary resistance.

PREVALENCE OF PRIMARY AS AGAINST ACQUIRED RESISTANCE

The distinction between primary and acquired resistance is based solely on whether a history of treatment with the relevant drug or drugs was elicited or not.

Table 4 shows the number and percentage of cases with primary and acquired resistance to the 3 drugs. Four cases with cultures resistant to isoniazid+ and 1 with a culture resistant to streptomycin or PAS for whom treatment histories were not available on any occasion have been excluded. Their exclusion is indicated in the column for those with primary resistance.

The prevalence of primary resistance to any of the 3 drugs (even assuming that all the 5 excluded cases had primary resistance) was almost the same at the 3 surveys (Table 4). It may be seen that the increase in the number of cases with resistance to isoniazid+ at Round III (shown in Table 2) is due entirely to increase in acquired resistance. Although this is not shown in the table, there was no significant difference between the sexes in the prevalence of primary or acquired resistance or of sensitive cultures at the first two rounds; but at the third round the proportion of females was significantly greater among those with primary resistance than among those with sensitive cultures ($P<0.01$).

Table 5 shows the number of cases with acquired or primary resistance and with sensitive cultures by X-ray status and by number of colonies grown on culture. Cases from all the 3 rounds have been pooled in the upper part of Table 5 and those from the first 2 rounds only in the lower part (if a case occurred at more than one survey it has been counted only at the initial survey). Further, the 5 cases excluded from Table 4 for whom history regarding treatment was not available have again been excluded. X-ray status is shown in 2 broad categories only: those who were X-ray cases and those who were not (as defined above under " Methods and Material ").

Among those with resistant cultures, the dominant figure in both parts of the table is of those who had acquired resistance to isoniazid+ and were X-ray cases and whose cultures grew 20 colonies or more. The figure in the upper part of the table could be rather artificial, being the result of the treatment given after Round II. It is to exclude the effect of this possibly artificial factor that the lower part is given, with data from the first 2 rounds only. Those with acquired resistance to isoniazid+, who were X-ray cases and whose cultures grew 20 colonies or more are still dominant; this remains true if a similar table is prepared from the data of the first round only (not shown). Therefore it is suggested that it is only those who are X-ray cases and whose

TABLE 5

DISTRIBUTION OF CASES WITH RESISTANT [a] AND SENSITIVE [b] CULTURES BY X-RAY STATUS AND BY NUMBER OF COLONIES GROWN ON CULTURE, IN 133 VILLAGES

Description	Resistance	X-ray cases [c]		Other than X-ray cases	
		No. of colonies grown on culture			
		⩾20	<20	⩾20	<20
Cases from all 3 rounds					
Cases with acquired resistance	To isoniazid+	38	5	1	2
	To streptomycin or PAS	1	1	—	—
Cases with primary resistance	To isoniazid+	6	5	6	8
	To streptomycin or PAS	5	4	2	6
Cases with sensitive cultures [d]		124	46	47	82
Cases from Rounds I and II only					
Cases with acquired resistance	To isoniazid+	25	1	—	1
	To streptomycin or PAS	1	1	—	—
Cases with primary resistance	To isoniazid+	6	4	4	5
	To streptomycin or PAS	4	4	—	2
Cases with sensitive cultures [d]		99	42	40	

[a] For cases with resistant cultures at more than one round, X-ray status and colony count have been considered only at the time of the first resistant culture.

[b] For cases yielding sensitive cultures at more than one round, X-ray status and colony count of the earliest round only have been considered.

[c] For definition, see " Methods and Material " (page 682).

[d] 16 cases (8 with ⩾20 colonies, 8 with <20 colonies grown on culture) which had not been X-rayed have been excluded.

cultures grow 20 colonies or more who usually develop resistance to isoniazid with the kind of treatment available in the area.

The following comparisons in Table 5 gave statistically significant results:

(*a*) The proportion of X-ray cases was significantly greater ($P<0.01$) among those with acquired resistance to isoniazid+ than among those with primary resistance to isoniazid+ or those with sensitive cultures, in both the upper and the lower parts of Table 5.

(*b*) The proportion of cases with 20 colonies or more grown on culture was significantly greater ($P<0.01$) among those with acquired resistance to isoniazid+ than among those with primary resistance to isoniazid+ or those with sensitive cultures, in both the upper and the lower parts of Table 5.

(*c*) The proportion of those with primary resistance was greater among those with resistance to PAS or streptomycin than among those with resistance to isoniazid ($P<0.01$ in both parts of Table 5).

(*d*) Among the non-X-ray cases, there was a significantly greater proportion of those with primary resistance than of those with acquired resistance ($P<0.01$ in both parts of Table 5).

(*e*) Among those whose cultures grew less than 20 colonies, again the proportion of those with primary resistance was greater than that of those with acquired resistance ($P<0.01$ in both parts of Table 5).

In addition all the 7 children below 15 years of age with resistant cultures had primary resistance; 3 of these had cultures resistant to streptomycin or PAS only. Thus there was a significantly higher frequency of primary resistance among the following: (*a*) those who were not X-ray cases, (*b*) those whose cultures grew less than 20 colonies, (*c*) those with resistance to streptomycin or PAS, (*d*) children.

Comparison of those with primary resistance with those with sensitive cultures did not show any significant difference regarding X-ray status or the number of colonies grown on culture.

CULTURE-POSITIVE CASES WITH NO EVIDENCE OF RADIOLOGICALLY ACTIVE DISEASE

It may be recalled that all cases with an X-ray reading of A or B were eligible for sputum examination (see " Methods and Material " above), not only at the particular round at which that reading was made, but also at any subsequent round, even though the X-ray picture might then be read as " normal " by both readers. The proportion of such cases in the 3 categories is now considered.

Cases with primary resistance

Of the 22 " other than X-ray cases " with primary resistance in the upper part of Table 5, 17 had not been assessed as being in X-ray category C or D at any round, nor had any of them sought any treatment at any round. These cases are detailed in the Appendix Table. Sputum cultures from some of these gave negative catalase and peroxidase tests (last column of the Appendix Table); 2 showed growth at room temperature. In a large number of these cases the number of colonies grown was small.

Follow-up of these cases showed that finding of cultures with primary resistance, once or more often, in such persons had no serious diagnostic or prognostic implication. A large number were culture-negative at a later round in the absence of any treatment.

Cases with sensitive cultures

Of the 129 " other than X-ray cases " with sensitive cultures in the upper part of Table 5, 103 had not been assessed as X-ray category C or D at the round at which the culture was positive and 53 had not been in those categories at any of the 3 rounds; there was no significant difference from those with primary resistance or in the proportion of those whose cultures grew less than 20 colonies. Among those with sensitive cultures there were 8 for whom cultures grew at room temperature; 9 of the cultures gave negative or doubtful reactions to peroxidase and catalase tests.

Cases with acquired resistance

The only 3 cases in the upper part of Table 5 with acquired resistance constituted a significantly smaller proportion of the total from all 3 rounds than the cases in the other 2 categories.

DISCUSSION

PREVALENCE AT THE 3 ROUNDS

There was no statistically significant difference in the prevalence of culture-positive cases at any of the 3 rounds. But, as seen in Table 4, the number of cases with acquired resistance to isoniazid alone increased significantly at Round III ($P<0.01$). Sputum-positive cases from Round I and all cases from Round II assessed as X-ray category D by both readers were offered treatment with isoniazid alone at Round II. Almost all such cases consumed the tablets for the first month, as these were given to them in their homes during the survey, but there was great irregularity in the subsequent collection of isoniazid, for which they had to go to the medical facility nearest to their home. Consequently, not only did the number of cultures resistant to isoniazid increase, but the degree of resistance to isoniazid was also considerably greater at Round III. The numbers of cultures showing a minimal inhibitory concentration of 50 μg/ml or more for the 3 rounds were 13, 12 and 29 respectively (not tabulated), the increase at the third round being highly significant ($P<0.01$).

Further, those with acquired resistance to isoniazid were mostly X-ray cases whose cultures grew 20 colonies or more (Table 5). It may be only among such probably more advanced cases that acquired resistance develops.

ARE CASES WITH ISONIAZID-RESISTANT CULTURES LESS INFECTIOUS TO THEIR HOUSEHOLD CONTACTS?

The infectivity of a case may be estimated by the number of tuberculin-negative household contacts who show evidence of new infection from one round to another, that is, by the incidence of new infections. Obviously only contacts tuberculin-tested on 2 occasions at least can be judged for new infections. It has been shown earlier that an increase of not less than 16 mm in the diameter of the tuberculin reaction from one test to another should be taken as evidence of new infection (Raj Narain et al., 1966). The

TABLE 6

PREVALENCE OF INFECTION AT THE TIME OF DIAGNOSIS AMONG HOUSEHOLD CONTACTS OF CASES WITH PRIMARY OR ACQUIRED RESISTANCE [a] TO ISONIAZID+ OR WITH SENSITIVE CULTURES IN 119 VILLAGES

Type of case		Round I	Round II	Round III
Primary resistance to isoniazid+	No. of contacts	28	30	33
	Percentage infected [b]	32.1	46.7	33.3
Acquired resistance to isoniazid+	No. of contacts	48	54	135
	Percentage infected [b]	62.5	64.8	62.9
Sensitive cultures	No. of contacts	471	362	457
	Percentage infected [b]	33.1	36.2	37.6

[a] Cases with resistant cultures for which history of treatment was not available have been excluded.

[b] Those with tuberculin reactions of 15 mm or more.

incidence of such new infections in different age-groups, separately from Round I to Rounds II and III and from Round II to Round III, was studied for cases with acquired or primary resistance and for those with sensitive cultures. The numbers of those at risk were small, especially for those with primary or acquired resistance. The incidence was generally, but not always, higher among the contacts of cases with sensitive cultures; in no case was the difference statistically significant; nor was there any consistent or significant difference between the infectivity of cases with acquired or primary resistance.

The prevalence of infection among household contacts of cases with primary or acquired resistance to isoniazid+ and of those with sensitive cultures *at the time of diagnosis* is shown in Table 6. Data from each round have been treated independently. Only those with tuberculin reactions of a diameter of 15 mm or more have been regarded as infected (Raj Narain, 1968).

The proportion of infected persons was consistently highest among households with a case with acquired resistance to isoniazid+ and the difference between these and household contacts of cases with sensitive cultures was highly significant for each of the 3 rounds (at the 99% level of confidence). The difference was also significant for different age-groups (not shown). The difference from household contacts of cases with primary resistance was statistically significant only for Rounds I and III and only at the 95% level of confidence. Between households with cases with primary resistance and those with sensitive cultures there was no significant difference.

The findings were similar when calculations were based on reactions of 10 mm or more as representing infection. Further, no significant difference was seen between household contacts of cases with acquired and with primary resistance to PAS or streptomycin or between either of these and contacts of cases with sensitive cultures.

This greater prevalence of infection at the time of diagnosis in households of cases with acquired resistance to isoniazid+ is also likely to be found in surveys conducted by other workers and could, by itself, be interpreted to mean that cases with resistant cultures are more infectious. In the light of the findings in this report such an interpretation would not be tenable, because: (*a*) the longer duration of sputum-positivity of cases with resistant strains, as shown in Fig. 3 and 4, was probably responsible for such a finding; and (*b*) the generally greater incidence of new infections in households containing cases with sensitive cultures, although statistically not significant, suggests that such cases might be more, and not less, infectious than cases with resistant cultures.

CHANGES IN SENSITIVITY STATUS FROM ONE ROUND TO ANOTHER

In Fig. 1, of the 9 cases with primary resistance at Round I, 3 had sensitive cultures at Round II and a fourth at round III. Among the 154 cases with

sensitive cultures, 1 of the 5 with primary resistance at Round II yielded a sensitive culture at Round III. In Fig. 2, of the 4 with primary resistance at Round II, 1 yielded a sensitive culture at Round III. Of cases with cultures resistant to streptomycin or PAS, 2 cases, with primary resistance at Round I, yielded sensitive cultures at Round III. Thus 8 cases, all with primary resistance, and a ninth described hereafter, showed a reversal from resistant cultures to sensitive cultures. All these cases were tuberculin-positive with a reaction size of 18 mm or more. Most grew only a few colonies; only 2 were direct-smear-positive.

It is true that resistant cultures may revert to sensitive cultures at a later date. Sensitive strains have been recovered after an interval from monkeys (Schmidt et al., 1958) and from guinea-pigs (Nouf-flard-Guy-Loe & Berteaux, 1966) infected with pure cultures of resistant strains, but in patients most sputum samples yielding resistant cultures contain both sensitive and resistant strains. It has been shown in Table 3 that one-third of those with primary resistance to isoniazid+ in one sputum specimen had sensitive cultures in the other specimen. The reversion of the 8 resistant cultures to sensitive cultures could therefore be a chance phenomenon, specimens containing both strains yielding sometimes sensitive and sometimes resistant cultures. But the occurrence of this only among those with primary resistance initially may not be due to chance alone. Changes of the opposite type also occurred; in 11 cases (as may be seen in Fig. 1 and 2) sensitive cultures showed primary resistance at a later round. Another possibility could be that atypical bacilli coexisted with typical bacilli. Whenever atypical bacilli grew, the cultures were resistant; when typical bacilli grew, the cultures were sensitive. This possibility is supported by the fact that among the 8 originally resistant cases, 4 subcultures of resistant strains showed growth at room temperature; 1 sensitive strain also did so. For 4 of these 8 cultures from Round I, the niacin test was not done. Among the 11 originally sensitive cases, 1 resistant and 1 sensitive strain grew at room temperature.

One of the 8 and one of the 11 were not X-ray cases at any of the 3 rounds. Coexistence of typical bacilli is not supported by the fact that only 1 of the 19 died and that some became culture-negative at later rounds without any treatment. Culture of typical bacilli from sputum could be expected to have a more serious prognostic implication. This finding is further discussed below.

The ninth case, referred to earlier, gives interesting findings. For this advanced case (in a 29-year-old woman) cultures were highly resistant to all 3 drugs at Round I (to PAS by "spot" sample only). At Round II cultures from both samples of sputum were sensitive to PAS, and for streptomycin the resistance ratio was reduced from 8 to 4. At Round III the cultures were sensitive to both PAS and streptomycin, but highly resistant to isoniazid. This case had been treated for a long time in a sanatorium with streptomycin, isoniazid and PAS before Round I. After the first round, the patient received irregular treatment with isoniazid and occasionally took streptomycin. After Round II treatment was with isoniazid only. It would appear that, with the discontinuance of treatment with PAS and streptomycin, resistance to those two drugs was lost. It is possible that resistance to a drug may be sustained only by continued treatment with it.

WERE SOME OF THE CULTURES THOSE OF ATYPICAL BACILLI?

In attempting to answer this question, several points must be taken into consideration.

Firstly, changes in sensitivity status occurred only among cases with primary resistance and it has been suggested above that some of these changes were possibly due to the coexistence of typical and a-typical bacilli in the same person. Also it has been pointed out that the presence of typical bacilli was doubtful as only one of 19 cases died and many became sputum-negative without any treatment.

Secondly, it is difficult to explain the occurrence of bacteriologically positive cases with no evidence of disease on X-ray examination (see page 692). Such cases are too large in number to be due only to errors, especially as the field work was well supervised. Further, the isolates are well distributed over the period of 5 years. The greatest frequency of errors in sputum collection or labelling would be expected within a village, as, on any one day, sputa were collected from one village only and were processed in the laboratory also on a day-to-day basis. However, the intra- and inter-village patterns of distribution do not support the possibility of errors due to confusion of sputum samples. Nor is the finding of such cases exclusive to the present series of surveys; earlier reports (Sikand & Raj Narain, 1958; Raj Narain et al., 1963; Roelsgaard et al., 1964) would appear to suggest that such cases are a more or less regular feature of surveys.

Thirdly, many differences between those with acquired resistance and those with primary resistance should be noted. As can be seen from the tables and figures:

(*a*) Mortality among those with acquired resistance was higher than among those with primary resistance (Fig. 1), although the difference was significant only for mortality of Round-I cases at Rounds II and III.

(*b*) A significantly larger percentage of those with primary than of those with acquired resistance at Round I became culture-negative in later rounds (Fig. 1).

(*c*) Only cases with primary resistance showed reversal to sensitivity at later rounds (Fig. 1).

(*d*) Cases with acquired resistance had been culture-positive much longer at the time of diagnosis (Fig. 3 and 4) and showed a much greater prevalence of infection among contacts than those with primary resistance (Table 6).

(*e*) A significantly larger proportion of those with acquired resistance were X-ray cases than of those with primary resistance or sensitive cultures (Table 5), while a large number of those with primary resistance had no evidence of radiologically active tuberculosis at any of the 3 rounds (Table 5 and Appendix Table).

(*f*) Most of the cases with resistance to PAS or streptomycin were those with primary resistance.

(*g*) Cases with primary resistance showed least agreement between findings by the 2 specimens of sputum (Table 3); the great majority were negative by the other specimen or yielded sensitive cultures.

It is difficult to explain such great differences between primary and acquired resistance if the disease in each case was due to *Myco. tuberculosis*. The disease in cases with primary resistance would appear to be very mild.

It is possible that errors in recording the history could vitiate the distinction between the primary and acquired resistance, but such errors in this unsophisticated population, with no motive for suppressing their correct history, would not appear to be likely.

It is this great contrast between acquired and primary resistance, the much greater lack of confirmation of culture results for one specimen by those for the other (Table 3) in cases with primary resistant cultures and the lack of confirmation of these culture results by any radiological evidence of disease that makes us suspect that some of the isolates with primary resistance could be of atypical bacilli in spite of their being niacin-positive.

If the cultures had been preserved, further identification tests would have confirmed or rejected the hypothesis. The need to preserve such isolates, obtained after great labour and cost, cannot be stressed too strongly. Such facilities must be developed.

Against this hypothesis is the fact that many findings in cases with primary resistance were similar to those in cases with sensitive cultures. For example, the proportions with no X-ray evidence of active disease or with cultures showing only a few colonies were not significantly different in the 2 groups. However, the higher proportion of cases with primary resistance among children and among those with resistance to PAS or streptomycin (as compared with resistance to isoniazid) should be noted. At Round III, the proportion of females among cases with primary resistance was significantly greater than among cases with sensitive cultures.

The significance of these differences is difficult to assess but they could be due to a proportion of the cases with primary resistance being really due to atypical bacilli. Unpublished data from one of our recent surveys (in Kancheepuram) showed that the prevalence of cases with cultures positive for atypical bacilli was similar in the two sexes while the prevalence of cases with cultures positive for *Myco. tuberculosis* in females was much less than that in males. The possibility that some of the isolates with primary resistance may be atypical bacilli has also been mentioned by Hobby (1963) and Hobby et al. (1965).

Another difficulty in accepting the hypothesis that some of the isolates with primary resistance were really atypical bacilli is the necessary further assumption that they were of the nonchromogenic type. The few isolates that developed pigment in the dark or after exposure to light have been excluded. Nonchromogenic niacin-positive mycobacteria have not been reported so far. Do such mycobacteria exist in this area? It may be of interest to add that 2 niacin-positive strains of *Myco. kansasii*, also not reported before, have been documented recently (Yue & Cohen, 1966). The area of our study is one with a high frequency of non-specific allergy.

PREVALENCE OF PRIMARY RESISTANCE

It has been shown in Table 4 that 7%–8% of the positive cultures had primary resistance to one or

another of the 3 drugs; the prevalence was more or less the same at the 3 surveys. Although based on different material—namely, patients reporting to clinics—the prevalence of primary resistance in the two national surveys in Great Britain, during 1955–56 and 1963, was 5.1% and 4.1% respectively (Fox et al., 1957; Miller et al., 1966), and in the area of the present surveys it was 15.4% (Gangadharam, 1967).

The relative proportion of those with primary resistance to the total of resistant cultures can be worked out from Table 4, and was 50%, 44% and 26% respectively at the 3 surveys. The high proportion of primary resistant cultures at the 3 rounds could be due to lack of facilities for treatment; it is only treatment that leads to acquired resistance. However, the possibility that some of the isolates were atypical bacilli has already been suggested as an explanation.

For practical purposes, it is suggested that, for estimating primary resistance, cases in whom the diagnosis of tuberculosis lacks certainty owing to lack of confirmation by X-ray examination or cases subcultures of whose sputa show growth at room temperature may be regarded as not being due to primary resistance. If cases with no X-ray evidence of active disease are excluded, the prevalence of primary resistance to any drug at the 3 rounds is reduced to 3.6%, 5.7% and 2.9% respectively. These are not significantly different from each other and may represent a truer version of the prevalence or primary resistance.

SUMMARY

Three tuberculosis prevalence surveys were carried out at intervals of 18 months in a sizeable random sample of villages in South India. The prevalence of cases with cultures positive for *Myco. tuberculosis* at the 3 survey rounds was 3.8‰, 4.0‰ and 3.7‰ respectively; of these 7%–8% had primary resistance to any of 3 drugs (isoniazid, streptomycin or PAS) at each of the 3 rounds, and 7.0%, 10.3% and 20.9% respectively had acquired resistance. The statistically significant increase in acquired resistance at the third round was due to an increase in cultures resistant to isoniazid only. The number of cultures highly resistant to isoniazid also increased significantly at Round III owing to irregular treatment with isoniazid after Round II. There was no difference in the prevalence of primary resistance at the 3 rounds.

Two specimens of sputum (" spot " and " overnight ") were examined from each person found to have any radiological abnormality. Sensitivity tests of these sputum cultures (where both specimens were positive) showed that agreement of results between specimens was very low for cases with primary resistance to any drug and also for cases with acquired resistance to PAS.

Mortality rates were significantly higher for cases with acquired resistance and lowest for those with primary resistance. A significantly larger number of cases with primary than with acquired resistance were culture-negative in subsequent rounds in the absence of treatment. It was only among those with primary resistance that reversal to sensitivity occurred at subsequent rounds. Other significant differences between those with primary and acquired resistance were that a larger proportion of the former were not X-ray cases and that they more often yielded only a small number of colonies on culture.

The great contrast between cases with acquired and those with primary resistance, the very mild disease among the latter, the absence of radiologically active disease among many of them, and the large number giving negative or sensitive cultures at a subsequent round leads us to suggest that a number of these cultures, although niacin-positive, were those of atypical bacilli. Further, it is suggested that, for estimates of prevalence of primary resistance in a community, cases with no radiologically active disease but with cultures showing primary resistance may be excluded, as among them even the existence of tuberculous disease is by no means certain.

At the time of diagnosis there was a much higher prevalence of infection in the household contacts of cases with acquired resistance than in those of cases with primary resistance or sensitive cultures, but the incidence of new infections among them on follow-up was not significantly different from that among the other two broad groupings by culture.

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RÉSUMÉ

Trois enquêtes sur la prévalence de la tuberculose ont été menées à 18 mois d'intervalle, au sud de l'Inde, dans un groupe relativement important de villages choisis par échantillonnage aléatoire. Lors des trois tournées effectuées par les enquêteurs, la prévalence des cas chez lesquels les cultures contenaient *Mycobacterium tuberculosis* a été respectivement de 3,8 pour 1000, 4,0 pour 1000 et 3,7 pour 1000; parmi ces malades, 7 à 8% présentaient une résistance primaire à l'un ou l'autre des trois médicaments (isoniazide, streptomycine ou PAS) lors de chacune des trois tournées, tandis que 7,0%, 10,3% et 20,9% respectivement présentaient une résistance acquise. L'augmentation statistiquement significative du nombre des résistances acquises observée lors de la troisième tournée s'explique par une augmentation du nombre des cultures résistantes à l'isoniazide seulement. Le nombre des cultures très résistantes à l'isoniazide avait également augmenté de façon significative lors de la troisième tournée, du fait de l'irrégularité avec laquelle le traitement par ce médicament avait été suivi après la deuxième tournée. En ce qui concerne la prévalence de la résistance primaire, on n'a enregistré aucune différence entre les trois enquêtes.

Pour chaque personne qui présentait une image radiologique anormale, on a examiné deux échantillons de crachats, l'un recueilli sur-le-champ et l'autre prélevé sur les crachats de la nuit. Les résultats des tests de sensibilité pratiqués sur les cultures ainsi obtenues (lorsque les deux échantillons étaient positifs) se sont rarement révélés concordants, aussi bien dans les cas de résistance primaire à l'un ou l'autre médicament que dans ceux de résistance acquise au PAS.

Les taux de mortalité ont été significativement plus élevés chez les malades présentant une résistance acquise; les plus faibles se rencontraient chez les malades présentant une résistance primaire. Au cours des tournées successives, on a vu apparaître, en l'absence de traitement, un nombre significativement plus grand de cultures négatives parmi les cas de résistance primaire que parmi ceux de résistance aquise, et ce n'est que parmi les premiers que se sont produits des retours à la sensibilité. D'autres différences significatives ont été notées entre les malades présentant soit une résistance primaire, soit une résistance acquise: il y avait une plus grande proportion d'images radiologiques normales chez les premiers que chez les seconds et il arrivait plus fréquemment que leurs cultures ne produisent qu'un petit nombre de colonies bacillaires.

Le profond contraste entre les malades qui présentent une résistance acquise et ceux qui présentent une résistance primaire, le caractère très atténué de la maladie chez ces derniers, l'absence chez beaucoup d'entre eux de signes radiologiques de tuberculose évolutive, ainsi que le grand nombre de sujets de ce groupe chez lesquels les cultures sont devenues négatives ou contenaient des bacilles sensibles au cours de la 2e ou de la 3e tournée, amènent les auteurs à penser que beaucoup de ces cultures, bien que donnant une réaction positive à l'épreuve de la niacine, étaient en fait des cultures de bacilles atypiques. Ils sont d'avis en outre que pour évaluer la prévalence de la résistance primaire dans une collectivité il ne faut pas tenir compte des cas chez lesquels les clichés radiologiques ne montrent pas de lésion évolutive même si les cultures indiquent une résistance primaire, car rien ne prouve que ces malades soient réellement atteints de tuberculose.

Au moment où le diagnostic de tuberculose a été posé, on a observé un nombre beaucoup plus élevé de cas d'infection tuberculeuse parmi les contacts familiaux des sujets présentant une résistance acquise que parmi les contacts des malades présentant une résistance primaire ou dont les cultures contenaient des bacilles sensibles. Cependant, lors des examens de contrôle, on n'a pas noté de différence significative de la fréquence des infections nouvelles parmi les contacts de malades porteurs de bacilles résistants et les contacts de malades porteurs de bacilles sensibles.

REFERENCES

Fox, W., Wiener, A., Mitchison, D. A. & Sutherland, I. (1957) *Tubercle (Edinb.)*, **38**, 71-84

Gangadharam, P. R. J. (1967) *Drug resistance in tubercle bacilli and its impact on chemotherapy and epidemiology of tuberculosis.* In: *Proceedings of the XXII All India Tuberculosis and Chest Diseases Workers' Conference, Hyderabad*, New Delhi, Tuberculosis Association of India, pp. 215-254

Hobby, G. L. (1963) *Amer. Rev. resp. Dis.*, **87**, 29-36

Hobby, G. L., Lenert, T. F., Maier, J. & O'Malley, P. (1965) *Amer. Rev. resp. Dis.*, **91**, 30-34

Middlebrook, G. & Dressler, S. H. (1954) *Amer. Rev. Tuberc.*, **70**, 1102-1103

Miller, A. B., Tall, R., Fox, W., Lafford, M. J. & Mitchison, D. A. (1966) *Tubercle (Edinb.)*, **47**, 92-103

Noufflard-Guy-Loe, H. & Berteaux, S. (1966) *Amer. Rev. resp. Dis.*, **94**, 540-552

Raj Narain (1968) *Tubercle (Edinb.)*, **49**, 92-103

Raj Narain, Geser, A., Jambunathan, M. V. & Subramanian, M. (1963) *Bull. Wld Hlth Org.*, **29**, 641-664; *Indian J. Tuberc.*, **10**, 85-116

Raj Narain, Nair, S. S., Chandrasekhar, P. & Ramanatha Rao, G. (1966) *Bull. Wld Hlth Org.*, **34**, 605-622

APPENDIX

17 CASES[a] WITH PRIMARY RESISTANT CULTURES BUT NO

Age at Round 1 (years)	Sex	X-ray readings			Culture results						Result of sensitivity tests: Isoniazid (MIC in μg/ml)					
		Round I	Round II	Round III	Round I		Round II		Round III		Round I		Round II		Round III	
					SP	OV	SP	OV	SP	OV	SP	OV	SP	OV	SP	OV
5	M	OO	AA	OO	—	—	N	+	N	N	—	—	—	>50	—	—
35	M	OO	AA	OO	—	—	N	N	1	N	—	—	—	—	5	—
11	M	OO	AO	OO	—	—	+	+	N	N	—	—	5	5	—	—
22	F	—	OA	OO	—	—	N	5	N	N	—	—	—	S	—	—
13	F	—	—	OA	—	—	—	—	N	1	—	—	—	—	—	>50
11	F	OO	OO	AO	—	—	—	—	2	1	—	—	—	—	S	S
55	F	—	—	OA	—	—	—	—	—	1	—	—	—	—	—	S
48	F	OO	OO	AO	—	—	—	—	5	N	—	—	—	—	S	—
37	M	BO	OO	—	N	1	N	N	D	D	—	>50	—	—	—	—
33	F	OO	BB	BB	—	—	1	N	N	N	—	—	>50	—	—	—
12	F	OCB	OO	OO	N	N	N	N	N	++	—	—	—	—	—	50
30	M	BB	OO	BB	N	1	N	N	N	N	—	50	—	—	—	—
30	M	OB	OO	OO	N	+	N	N	—	—	—	>50	—	—	—	—
65	F	BA	OO	AO	N	+	N	N	N	—	—	50	—	—	—	—
45	F	OB	OO	OO	1	3	N	1	2	—	S	S	—	S	S	—
7	F	—	—	OB	—	—	—	—	+	6	—	—	—	—	S	S
12	F	OO	OO	AB	—	—	—	—	+	N	—	—	—	—	S	—

[a] In no case was there a history of previous treatment.

[b] Abbreviations used:

General: SP = Spot sample of sputum. OV = Overnight sample of sputum. — = Examination not done.

X-ray readings: O = No abnormality. A = Probably non-tuberculous. B = Probably tuberculous but inactive. C = Probably tuberculous, possibly active. D = Probably tuberculous and active.

X-ray readings of the two readers are represented by a capital letter for each, e.g., CC. The third letter, where shown, represents the reading of the third reader (see text).

REFERENCES *(continued)*

Rao, K. P., Nair, S. S., Naganathan, N. & Ramanatha Rao, G. (1966) *Indian J. Tuberc.*, **13**, 147-157

Roelsgaard, E., Iversen, E. & Bløcher, C. (1964) *Bull. Wld Hlth Org.*, **30**, 459-518

Schmidt, L. H., Grover, A. A., Hoffman, R., Rehm, J. & Sullivan, R. (1958) *The emergence of isoniazid-sensitive bacilli in monkeys inoculated with isoniazid-resistant strains.* In: *Transactions of the Seventeenth Conference on the Chemotherapy of Tuberculosis*, Washington, D.C., US Veterans Administration, pp. 264-269 (quoted by Noufflard-Guy-Loe & Berteaux, 1966)

Šerman, A. Š. (1963) *Probl. Tuberk.*, **41**, No. 4, pp. 3-6 (abstracted in *Excerpta med. (Amst.), Sect. XV*, 1964, **17**, 254)

Sikand, B. K. & Raj Narain (1958) *Radiological problems of tuberculosis as revealed by the national survey.* In: Tuberculosis Association of India, *Proceedings of the Fourteenth All India Tuberculosis and Chest Diseases Workers' Conference, held in Madras...*, New Delhi, Tuberculosis Association of India, pp. 75-84

Yue, W. Y. & Cohen, S. S. (1966) *Amer. Rev. resp. Dis.*, **94**, 447-449

TABLE

RADIOLOGICAL EVIDENCE OF ACTIVE PULMONARY TUBERCULOSIS [b]

Results of sensitivity tests												" Abnormal " identification-test results
Streptomycin (resistance ratio)						PAS (resistance ratio)						
Round I		Round II		Round III		Round I		Round II		Round III		
SP	OV	SP	OV	SP	OV	SP	OV	SP	OV	SP	OV	
—	—	—	S	—	—	—	—	—	S	—	—	Catalase test, doubtful; peroxidase test, mixed [c]
—	—	—	—	4	—	—	—	—	—	S	—	
—	—	S	S	—	—	—	—	S	S	—	—	Peroxidase test, mixed [c] in both cultures
—	—	—	S	—	—	—	—	—	32	—	—	
—	—	—	—	—	S	—	—	—	—	—	16	Catalase and peroxidase tests, negative
—	—	—	—	16	16	—	—	—	—	S	S	
—	—	—	—	—	8	—	—	—	—	—	S	Peroxidase test, mixed [c]
—	—	—	—	S	—	—	—	—	—	32	—	
—	S	—	—	—	—	—	S	—	—	—	—	Catalase and peroxidase tests, negative
—	—	S	—	—	—	—	—	>32	—	—	—	Growth at room temperature
—	—	—	—	—	S	—	—	—	—	—	S	Catalase and peroxidase tests, negative
—	S	—	—	—	—	—	64	—	—	—	—	Niacin test not done
—	>4	—	—	—	—	—	>8	—	—	—	—	Luxuriant growth at room temperature; catalase and peroxidase tests, negative
—	S	—	—	—	—	—	16	—	—	—	—	Peroxidase test, negative
S	S	—	S	S	—	S	S	—	S	8	—	
—	—	—	—	16	16	—	—	—	—	S	S	
—	—	—	—	16	—	—	—	—	—	S	—	

Culture results: + = Culture growing more than 20 colonies. Where less than 20 colonies grew, the number is shown in Arabic numerals.
N = Culture negative.
D = Dead.

Sensitivity tests: S = Sensitive to all 3 drugs.

[c] A " mixed " result in the peroxidase test indicates that some colonies were positive and some negative.