Recurrence of tuberculosis in a low incidence setting

Claudia C. Dobler, A.B. Hamish Crawford, Peter J. Jelfs, Gwendolyn L. Gilbert, Guy B. Marks

1 Respiratory Department, Liverpool Hospital, Sydney, New South Wales, Australia
2 Centre for Infectious Diseases and Microbiology, Institute of Clinical Pathology and Medical Research, Westmead Hospital, Sydney, New South Wales, Australia

Corresponding author, requests for reprint:

Claudia Dobler
Address:
Liverpool Hospital
Respiratory Department
Clinical Services Building, Level 1
Elizabeth Street
Liverpool NSW 2170
Australia

Phone: +61 0406621292
Fax: +61 (02) 98294102
Email: ccdobler@yahoo.com

Short title for the manuscript: Recurrent tuberculosis
ABSTRACT

Recurrence of active tuberculosis after treatment of an initial disease episode can be due to endogenous reactivation or exogenous reinfection.

Cases of recurrent tuberculosis in the Australian state of New South Wales between 1994 and 2006 were identified by data linkage analysis with confirmatory review of case notes. We included patients with more than one culture positive disease episode during that time period who had completed treatment for the initial disease episode. Genotyping of *Mycobacterium tuberculosis* was used to determine whether recurrence was likely to be due to reactivation or reinfection.

There were 5723 tuberculosis notifications between 1994 and 2006, of which 3731 were culture positive. Fifteen patients (0.4%) had recurrent culture positive disease over an average 5.7 years of follow-up (crude annual incidence, 71/100,000). Recurrent tuberculosis was attributable to reactivation (indistinguishable strains) in 11 (73%) cases and to reinfection (different strains) in 4 cases (27%).

In a low incidence setting for tuberculosis, a control program incorporating directly observed therapy for active disease results in a very low rate of recurrent tuberculosis over a long period of follow-up. Reinfection is less likely than reactivation, but still contributes significantly to the number of cases with recurrent disease.

**Keywords:**
directly observed therapy, genotyping, reactivation, recurrence, reinfection, tuberculosis epidemiology
INTRODUCTION

Rates of recurrence of tuberculosis (TB) reflect the long term efficacy of tuberculosis treatment, the effectiveness of the tuberculosis control strategy, the underlying incidence of tuberculosis in the community, and the immune status of the community. Rates of recurrent tuberculosis in settings with a low to intermediate incidence of tuberculosis are reported to range from 1.2-7% [1-4]. Recurrence of active tuberculosis after treatment of an initial disease episode may be due to endogenous reactivation with the same strain of *Mycobacterium tuberculosis* or to exogenous reinfection with a different strain. In countries with a low or intermediate incidence of tuberculosis recurrence is usually suspected to be due to reactivation rather than reinfection [1-8]. In contrast, in high-incidence countries reinfection seems to be a major cause of recurrent disease [9, 10]. HIV-infection increases the risk of recurrent disease because HIV-positive individuals infected with *Mycobacterium tuberculosis* rapidly progress to disease. Furthermore, there is some evidence that they may have a higher risk of developing infection after exposure to *Mycobacterium tuberculosis* [11]. Traditionally it was assumed that recovery from an episode of tuberculosis at least partially protects an individual against subsequent tuberculosis due to reinfection with another strain. However, a recent study from South Africa has suggested that people who had tuberculosis once are at increased risk of having another episode of disease [9].

In New South Wales, Australia, the incidence of tuberculosis and HIV infection are both low. Over 80% of cases of TB occur in people born overseas, presumably representing reactivation of latent TB infection. Virtually all patients are treated with directly observed therapy (DOT). We wished to establish the incidence of recurrent disease and the relative contributions of reactivation and reinfection in this setting.
PATIENTS AND METHODS

Setting
Our study was conducted in New South Wales (NSW), Australia’s most populous state, with an approximate population of 6,800,000 (almost one third of Australia’s total population). The incidence of tuberculosis in New South Wales is 6.5/100,000 [12]. Tuberculosis treatment is given free of charge through the local chest clinics. Chest clinics are public hospital-associated outpatient centres that are specialised in the management of tuberculosis.

Study population
Tuberculosis is a notifiable disease in New South Wales (NSW). There is evidence that the NSW tuberculosis notifications database is a complete, and hence sensitive, record of cases of tuberculosis in NSW [13]. We reviewed all tuberculosis notifications between 1994 and 2006 in this database. Data linkage within this database was used to identify cases that had more than one TB notification in that time period. We included patients in our analysis if, between 1994 and 2006, they had more than one disease episode, both of which were culture positive, and they had completed treatment for the initial episode of tuberculosis.

Data linkage, data analysis
Records with identical date of birth within the NSW tuberculosis notification database were identified via computer-generated search. These pairs of records were then manually checked for similar names, sex, country of origin, date of arrival in Australia. Notification dates in different records for the same patient had to be at least 6 months apart, otherwise we assumed that they were duplicate notifications for the same episode, e.g. in cases where patients were transferred to another chest clinic during treatment, and both institutions notified the patient. Cases with multiple notifications that were at least 6 months apart were further analysed by
accessing clinical records. Duplicate notifications for the same episode or apparent recurrences with culture negative disease in any disease episode were excluded. Verification of culture results (by accessing the clinical records) was sought for all cases of recurrence, even if the database stated negative cultures. For the whole cohort of TB notifications, we accepted a case to be culture positive if either or both of the following were recorded:

1. Positive culture for *Mycobacterium tuberculosis* from a dated specimen
2. “Culture” stated as the disease identifying method

Cases of multi drug resistant tuberculosis (MDR-TB) were identified from the records of the NSW Mycobacterium Reference Laboratory at the Institute of Clinical Pathology and Medical Research, Westmead Hospital, New South Wales, Australia.

**Treatment**

Patients were treated in accordance with NSW Department of Health Tuberculosis Guidelines which require directly observed therapy (DOT) for a minimum period of 6 months using initially isoniazid, rifampicin, pyrazinamide and ethambutol for a minimum of 2 months and rifampicin and isoniazid for the subsequent 4 months in fully sensitive disease [14]. All patients were directly observed taking all doses of their medication at the chest clinics or in their home, except that patients on daily treatment were not observed taking their weekend doses. Treatment regimens were no different whether a patient received medication at the chest clinic or at home. Cases of MDR-TB were treated with DOT using second-line treatment over a period of 12-24 months.

**Follow-up period**

Patients were usually subject to active radiological surveillance of at least 2 years post treatment cessation. For this analysis the average period of time from cessation of treatment to December 2006, when the linkage analysis was performed, was 5.7 years.
**Genotyping of M. tuberculosis isolates**

Isolates of *Mycobacterium tuberculosis* for genotyping were obtained from patients with culture positive recurrent tuberculosis for both disease episodes. One *Mycobacterium tuberculosis* isolate from each disease episode from each patient with culture positive recurrent tuberculosis was used for genotyping. Genotyping was performed at the time of the second disease episode or retrospectively for this study from stored culture material at the Institute of Clinical Pathology and Medical Research, Westmead Hospital, New South Wales, Australia. Cross-contamination was excluded by appropriate molecular laboratory techniques, that is, the use of positive, negative and contamination controls and by testing unrelated isolates in the same batch. DNA fingerprinting was performed by using a combination of three methods namely spoligotyping [15], mycobacterial interspersed repetitive unit (MIRU) typing [16] and IS6110 restriction fragment length polymorphism (RFLP) typing [17]. Isolates were classified as distinct if they were clearly different by any of these three methods. RFLP was usually only performed for isolates that were indistinguishable after spoligotyping and MIRU typing. Disease attributable to reinfection was defined as a recurrent disease episode in which the strain had a genotype distinct from that which was isolated in the initial episode. Recurrences in which the strains from the two episodes were indistinguishable were attributed to reactivation.

**Data analysis**

Incidence rates were calculated per person-years of follow-up. Ninety-five percent confidence intervals around the estimated rates were calculated according to the asymptotic method of Fleiss [18]. The Kaplan-Meier method was used to construct a survival curve, with survival defined as being free of active TB.
Ethical Considerations

Ethical approval was obtained from the New South Wales Population and Health Services Research Ethics Committee, Australia.

Preliminary data from the study (results from one chest clinic in New South Wales) have been reported previously [19].
RESULTS

Description of the Cohort

There were 5723 TB notifications between 1994 and 2006 of which 3731 were culture positive (Figure 1). Characteristics of the cohort patients are shown in Table 1. The average age of the study population was 46.7 (standard deviation 21.1) years and 1935 (51.9%) of the culture positive cohort were male. Information on the country of birth was available for 3608 of these cases. Of these, 509 (14.1%) were Australian-born. The majority of the overseas born cases originated from Asia.

Incidence of Recurrent Tuberculosis

Fifteen patients (0.40%; 95% confidence interval [95% CI], 0.23 to 0.68) had recurrent culture positive disease after completion of treatment for an initial disease episode over an average 5.7 years of follow-up. The recurrent episodes occurred between 1.5 and 100 months (median 17 months) after cessation of treatment for the initial disease episode (Figure 2). The incidence of recurrent culture positive tuberculosis was 70.7 (95% CI, 16.6 to 241.6) per 100,000 person-years of follow-up. Table 1 shows the characteristics of TB cohort as a whole and also the cases with recurrent tuberculosis. Due to the low number of cases of recurrent TB no formal statistical tests have been performed. Paired DNA fingerprints were available in all 15 cases. Recurrent tuberculosis was attributable to reactivation (indistinguishable strains) in 11 cases and to reinfection (different strains) in 4 cases. (Table 2 summarises clinical and molecular typing data).

Of the 51 cases with MDR-TB in the initial or only episode, only one had recurrent disease, and that proved to be due to reinfection with a strain that was resistant only to isoniazid (patient 2 in Table 2). Of the remaining 14 cases with recurrent disease, 3 had MDR-TB in
the second disease episode, of which 2 were due to reactivation and one was due to reinfection with an MDR-TB strain, having been infected with a fully sensitive strain initially. Among all tuberculosis notifications, including culture negative cases, there were 26 recurrent cases out of 5723 notifications (0.45%). Hence, the estimate of the recurrence rate was not altered by limiting the analysis to culture positive cases only.

** Reactivation **

Reactivation occurred in 11 cases (0.29%; 95% CI, 0.15 to 0.54). Time to recurrence for reactivation cases ranged from 1.5 to 100 months (median 21.5 months) after cessation of treatment for the initial disease episode. The annual incidence of reactivation was 51.9 (95% CI, 8.6 to 213.3) per 100,000 person-years of follow-up. Of the 11 cases with reactivation, two had MDR-TB in the second disease episode. One of these patients (patient 10 in table 2) had initially a fully sensitive organism. She was diagnosed with HIV/AIDS at the time of presentation with the first episode of active tuberculosis. Severe diarrhoea, possibly related to HIV/AIDS, was an ongoing issue in this patient, and might have altered the absorption of the TB medication (no drug levels were performed). The patient had recurrent disease just 6 weeks after completion of treatment for the first disease episode. In the other case with MDR-TB on reactivation (patient 12 in Table 2), the initial episode was attributable to an organism that was resistant to isoniazid. He was treated with isoniazid, rifampicin, pyrazinamide and ethambutol for 9 months, and presented with reactivated disease 10 months after cessation of treatment for the first episode.

** Reinfection **

Reinfection occurred in 4 cases (0.11%; 95% CI, 0.03 to 0.29) of all culture positive cases over the period of follow-up. Time to recurrence for reinfection cases ranged from 8 to 56.5
months (median 16.3 months) after cessation of treatment for the initial disease episode. The crude annual incidence of recurrent disease due to reinfection was 18.9 (95% CI, 0.3 to 160.9) per 100,000 person-years, which is four and a half times the incidence rate of new culture positive tuberculosis (4.2/100,000).

Of the 4 cases with recurrent tuberculosis due to reinfection at least 3 (patients 1–3 in Table 2) had visited countries with a high incidence of tuberculosis following treatment of their initial episode in Australia. It is assumed reinfection occurred during those visits. One of these patients (patient 3 in Table 2) had received an autologous bone marrow transplant for multiple myeloma and was on treatment with alpha-interferon prior to both episodes of tuberculosis. Although she was born in Australia she had visited a high-incidence country for tuberculosis between completion of treatment for the first disease episode and episode of recurrent disease. The number of reinfection cases was too small for statistical analysis of risk factors for exogenous reinfection.

**Molecular typing**

The differences we found in strains attributed to reinfection were of significance beyond consideration of a mutational event. Only a quarter of our patient isolates had an identical spoligotype but in 5/12 of the MIRU loci in these isolates, the repeat numbers differed by 1-2.

Isolates from two patients had been typed by RFLP (the only method available in 1997) at the time of their recurrent disease. In both cases the paired isolates were indistinguishable, indicating reactivation. These isolates were no longer available for spoligotyping and MIRU typing. There was a range of spoligotype families among the remaining isolates, of which the commonest was Beijing (Table 2); two of three MDR isolates for which spoligotyping results were available were Beijing. One patient had separate episodes of infection with two different
Beijing strains (patient 2 in Table 2). In general, spoligotypes reflected the patients’ countries of origin.
DISCUSSION

This study shows that in this population, living in a setting with a low incidence of TB and low prevalence of HIV infection and managed with directly observed therapy, there is a very low incidence of recurrent tuberculosis. The annual incidence of recurrent tuberculosis, over an average period of 5.7 years of follow-up, was 71/100,000. Only 0.4% of all culture positive notifications were for recurrent disease. The proportion of cases that recurred in New South Wales appears to be substantially lower than that previously reported in comparable studies of treatment outcomes in low to intermediate incidence populations, ranging from 1.2-7% [1-4], the majority of which did not utilise DOT.

Although the number of patients with MDR-TB is small, our outcomes of DOT with second-line drugs are encouraging. Indeed, none of the patients treated for MDR-TB in a first disease episode had reactivation disease with the initial organism during follow-up.

Recurrent TB is most likely to be with drug-susceptible organisms. Our finding, that only two of eleven cases with reactivation had MDR-TB in the second disease episode is consistent with the findings of others [1, 2, 8, 21, 22]. That finding supports our current practice that in cases of recurrent TB, patients are not routinely started on second-line treatment.

In countries with a low or intermediate incidence of tuberculosis recurrent TB is mainly attributable to reactivation. In this setting the proportion of recurrent cases attributable to reinfection has been reported to range from 4 to 33% [1-7]. In contrast, in high-incidence countries reinfection may account for up to 77% of cases of recurrent disease [9, 10]. While the rate of reinfection reflects the incidence of tuberculosis in the area, a low rate of reactivation is a consequence of treatment efficacy, both in terms of optimal drug therapy and
patient compliance. The low rate of reactivation in our study has led to a relatively higher proportion of recurrent cases that are attributable to reinfection (27%). The relatively high incidence of recurrent disease due to reinfection in our population, relative to the very low incidence of new-onset culture positive disease, may also relate to high rates of overseas travel among Australians, particularly those who were born overseas. Travellers to areas of high tuberculosis endemicity have been shown to be at substantial risk of tuberculosis infection and disease [23, 24]. Of the 4 cases with reinfection at least 3 had stays in a country with a high incidence of tuberculosis following their initial treatment in Australia.

A strength of our study is the use of data from a TB control program, which is more representative of real life practice than is the experience from clinical trials. However, there are also limitations associated with the use of these data. The cohort was not limited to patients who met the criterion for cure as defined by the World Health Organization [25], but also included patients that were classified as having completed treatment. This is because follow-up culture results were not routinely documented. There were two cases of recurrence within 6 months after completion of treatment for the first episode. One case with reactivation after 6 weeks (patient 10 in Table 2) had evidence of sputum smear conversion during treatment, but no sputum examination was performed in the last month of treatment. The case with reactivation after 5.5 months (patient 15 in Table 2) was initially diagnosed on bronchial lavage specimens rather than sputum, and no follow-up bronchoscopies were performed routinely. Hence, we cannot exclude the possibility that these two cases represent primary treatment failure rather than recurrent disease. A further limitation of the study is that we are unable to account for recurrent cases that occurred among patients who left New South Wales during the follow-up period. If we assume a similar rate of departure from NSW as was found in a random sample of 200 refugees who arrived in Sydney, New South Wales, between 1984 and 1994 [13], we would estimate that 15% of the cohort left New South Wales during the
average follow-up period of 5.7 years. However, our cohort includes many longer term residents of NSW and this estimate is likely to be an over-estimate of the rate of out-migration. Hence, this is likely to have only a small impact on the estimates.

We can only speculate about the reasons for the low reactivation rate in our study population, as the lack of a control group does not allow us to draw any definite conclusions. One possible explanation is that treatment for tuberculosis was administered by units specialized in the management and treatment of tuberculosis disease under DOT. We think that the consistent application of DOT, as strongly advocated by the World Health Organisation in treatment programs for active tuberculosis, may at least partially be responsible for the good treatment outcomes. Nevertheless, we are aware that a number of prospective randomized trials in high-incidence populations have failed to demonstrate that DOT, per se, results in improved outcomes in the treatment of active TB [26, 27]. Another possible explanation for the low reactivation rate may be the concentration of TB treatment in specialized centres which assures that standardized treatment regimens of proven efficacy, using the recommended doses and for the recommended duration are prescribed. Besides supervision, specialized centres can offer appropriate treatment support, which is especially important in non-compliant patients and patients experiencing possible side effects of TB treatment. In low incidence settings, concentration of all TB treatment in specialized centres is also likely to improve the expertise of the health professionals that are part of a treating team. This may also improve the management of MDR-TB patients. Finally the fact that in New South Wales all TB treatment is given free of charge, independent of insurance and residency status, may have improved compliance of patients with the treatment regimen and hence led to good outcomes.
Although the incidence of reinfection disease (18.9/100,000) was low, it was still four and a half times the incidence rate of new culture positive tuberculosis (4.2/100,000). A study from South Africa, a high-incidence country for tuberculosis, has reported a similar finding [9]. However, the incidence of reinfection among foreign-born persons (17.6/100,000) was similar to the overall incidence of culture positive tuberculosis among the foreign-born persons in New South Wales (16.2/100,000). This is a more realistic comparison because this is the population in which 85% of cases of TB occur. Thus, within this population, reinfection occurs at a similar rate to initial infection.

In our cohort, 507 of 3731 patients (13.6%) had HIV status assessed, of which 64 (12.6%) were HIV positive. More recently there have been efforts in New South Wales to introduce routine HIV testing in all TB cases. In 2005, among 393 cases in which HIV status was assessed (37% of all notified cases that year), only 9 (2.3%) were HIV positive. These findings suggest that in earlier years when significantly fewer patients were tested for HIV, testing was biased towards patients at higher risk for HIV. Although the numbers of cases with recurrent TB that were tested for HIV were very small, our data shows that HIV positive status tends to be more prevalent in patients with recurrent disease than in the whole cohort of TB notifications (see Table 1).

Recently developed molecular typing methods have made it relatively easy to distinguish reinfection from reactivation and the PCR-based methods (spoligotyping and MIRU typing) can be performed on nonviable isolates or stored DNA samples. For RFLP typing, the organism must be viable and cultured for a considerable period to generate the relatively large amount of bacterial DNA required. Although the combination of three typing methods provides the highest level of discrimination, isolates that are indistinguishable by a combination of spoligotyping and MIRU, in the contact of recurrent disease, are almost
certain to belong to the same strain. Spoligotyping alone is less discriminatory, especially in populations in which the Beijing family is relatively common. One patient (patient 2 in Table 2) had two episodes of infection with Beijing family strains that were indistinguishable by spoligotyping but easily distinguished by MIRU typing. The most common genotype in NSW is Beijing (26%) [28]. Of the eleven cases with reactivation, nine had information on spoligotype family/strain available. Two of them (22.2%) were cases with Beijing family strains, which reflects the prevalence of the Beijing genotype in NSW.

There is some uncertainty about the classification of cases as attributable to reinfection and reactivation on the basis of strain typing. For example, one of the 11 patients (patient 9 in Table 2) who had recurrent disease attributable to the same strain as the initial disease episode, was part of a known cluster of tuberculosis cases that was confirmed by genotyping and epidemiological linkage. Members of this disease cluster had continuing contact with each other at the time the second disease episode occurred [29]. Hence, it is possible that in this case the second disease episode was in fact due to exogenous reinfection with the same strain rather than reactivation. On the other hand, there is also a theoretical possibility that cases classified as reinfection due to different strains actually had a simultaneous infection with different strains at the time of the initial episode. If only one genotype was detected initially and the other genotype was detected at the time of recurrence this would have been falsely classified as being attributable to reinfection [30]. Although the likelihood of this is low, this possibility cannot entirely be excluded.

We conclude that in a low incidence area for tuberculosis, where virtually all cases of active disease are managed by DOT in specialized centres, the rate of tuberculosis recurrence is extremely low. Furthermore, disease due to reactivation is more likely to be drug sensitive than not. In this setting with high rates of travel to endemic countries, reinfection contributes
over a quarter of all cases of recurrence and is likely to be acquired during visits to high-incidence countries. Patients, who have been effectively treated for active tuberculosis remain susceptible to reinfection disease, especially when travelling to countries with a high incidence of tuberculosis. MDR-TB is no more likely to relapse than fully sensitive disease, possibly suggesting that treatment through centres that are specialized in the management of tuberculosis and treat every case of active disease with DOT results in successful control of MDR-TB in a low incidence setting.

ACKNOWLEDGMENTS

We thank the National Health and Medical Research Council Centre of Clinical Research Excellence in Respiratory and Sleep Medicine for providing funding for Claudia Dobler. The funding is independent of this specific study. We thank the New South Wales Department of Health for granting access to the tuberculosis notification data for database linkage, and the area tuberculosis coordinators of NSW for helping access clinical records.
Table 1 Characteristics of TB notifications cohort and cases of recurrent tuberculosis

<table>
<thead>
<tr>
<th></th>
<th>Cohort, n=3731</th>
<th>Recurrent TB cases, n=15</th>
<th>Cases with exogenous reinfection, n=4</th>
<th>Cases with endogenous reactivation n=11</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age yrs</strong></td>
<td>46.7(±21.1)</td>
<td>48.5 (±22.4)</td>
<td>31.3 (±15.5)</td>
<td>54.8 (±21.6)</td>
</tr>
<tr>
<td><strong>Sex</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>male</td>
<td>1935 (51.9%)</td>
<td>8 (53.3%)</td>
<td>0</td>
<td>8 (72.7%)</td>
</tr>
<tr>
<td>female</td>
<td>1796 (48.1%)</td>
<td>7 (46.7%)</td>
<td>4 (100%)</td>
<td>3 (27.3%)</td>
</tr>
<tr>
<td><strong>Country of origin</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Information available</td>
<td>3608</td>
<td>15</td>
<td>4</td>
<td>11</td>
</tr>
<tr>
<td>Australia</td>
<td>509 (14.1%)</td>
<td>3 (20%)</td>
<td>1 (25%)</td>
<td>2 (18.2%)</td>
</tr>
<tr>
<td>Overseas</td>
<td>3099 (85.9%)</td>
<td>12 (80%)</td>
<td>3 (75%)</td>
<td>9 (81.8%)</td>
</tr>
<tr>
<td><strong>HIV status</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Assessed</td>
<td>507</td>
<td>6</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td>positive</td>
<td>64 (12.6%)</td>
<td>2 (33.3%)</td>
<td>1 (50%)</td>
<td>1 (25%)</td>
</tr>
<tr>
<td><strong>Acid-fast direct smear†</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Information available</td>
<td>3261</td>
<td>14</td>
<td>4</td>
<td>10</td>
</tr>
<tr>
<td>Positive</td>
<td>1427 (43.8%)</td>
<td>10* (71.4%)</td>
<td>4* (100%)</td>
<td>7* (70%)</td>
</tr>
<tr>
<td>Negative</td>
<td>1834 (56.2%)</td>
<td>4* (28.6%)</td>
<td>0*</td>
<td>3* (30%)</td>
</tr>
<tr>
<td><strong>Site of disease</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Information available</td>
<td>3606</td>
<td>15</td>
<td>4</td>
<td>11</td>
</tr>
<tr>
<td>Pulmonary only</td>
<td>2137 (59.3%)</td>
<td>12* (80%)</td>
<td>3* (75%)</td>
<td>9* (81.8%)</td>
</tr>
<tr>
<td>Pulmonary and other site</td>
<td>257 (7.1%)</td>
<td>3* (20%)</td>
<td>1* (25%)</td>
<td>2* (18.2%)</td>
</tr>
<tr>
<td>Extra-pulmonary only</td>
<td>1212 (33.6%)</td>
<td>0*</td>
<td>0*</td>
<td>0*</td>
</tr>
<tr>
<td>MDR-TB</td>
<td>51 (1.4%)</td>
<td>1* (6.7%)</td>
<td>1* (25%)</td>
<td>0*</td>
</tr>
</tbody>
</table>
* at time of first disease episode

† at least on one specimen of the following: sputum, bronchial washings/lavage, lymph node fine needle aspiration
Table 2 Characteristics of cases with recurrent TB and molecular typing of paired *Mycobacterium tuberculosis*

<table>
<thead>
<tr>
<th>Case, episode</th>
<th>Patient’s country of birth</th>
<th>HIV status</th>
<th>Months to recurrence</th>
<th>Diagnostic site</th>
<th>Smear results</th>
<th>Year</th>
<th>Drug susceptibility</th>
<th>Spoligotype</th>
<th>Spoligotype family/strain</th>
<th>MIRU type††</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Reinfections (different strains)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1a Vietnam</td>
<td>Negative</td>
<td>15.5</td>
<td>Lung only, Lung only</td>
<td>Bronchial washings+, sputum +</td>
<td>2000</td>
<td>Susceptible</td>
<td>MDR</td>
<td>77777774413771</td>
<td>EIA4</td>
<td>234321223543</td>
</tr>
<tr>
<td>lb Vietnam</td>
<td>Unknown</td>
<td>15.5</td>
<td>Lung only, Lung only</td>
<td>Sputum +</td>
<td>2001</td>
<td>Susceptible</td>
<td>MDR</td>
<td>00000000003771</td>
<td>Beijing</td>
<td>223425173434</td>
</tr>
<tr>
<td>2a Vietnam</td>
<td>Unknown</td>
<td>8</td>
<td>Lung plus lymphatic site, Lung only</td>
<td>Lymph node+, sputum –</td>
<td>2002</td>
<td>MDR</td>
<td>INH-R</td>
<td>00000000003771</td>
<td>Beijing</td>
<td>222325173543</td>
</tr>
<tr>
<td>2b Vietnam</td>
<td>Unknown</td>
<td>8</td>
<td>Lung only, Lung only</td>
<td>Sputum –</td>
<td>2004</td>
<td>INH-R</td>
<td>MDR</td>
<td>00000000003771</td>
<td>Beijing</td>
<td>223326171531</td>
</tr>
<tr>
<td>3a Australia</td>
<td>Unknown</td>
<td>17</td>
<td>Lung only, Lung only</td>
<td>Sputum, Sputum +</td>
<td>1996</td>
<td>Susceptible</td>
<td>Susceptible</td>
<td>777777777720771</td>
<td>Beijing</td>
<td>223324173533</td>
</tr>
<tr>
<td>3b Australia</td>
<td>Unknown</td>
<td>17</td>
<td>Lung only, Lung only</td>
<td>Sputum –</td>
<td>1998</td>
<td>Susceptible</td>
<td>Susceptible</td>
<td>477777777413071</td>
<td>EA13/“Indian”</td>
<td>254326223534</td>
</tr>
<tr>
<td>4a Somalia</td>
<td>Positive</td>
<td>56.5</td>
<td>Lung only, Lung only</td>
<td>Sputum +, Sputum +</td>
<td>1999</td>
<td>Susceptible</td>
<td>Susceptible</td>
<td>777777777720771</td>
<td>Haarlem 3</td>
<td>225323153323</td>
</tr>
<tr>
<td>4b Somalia</td>
<td>Positive</td>
<td>56.5</td>
<td>Lung only, Lung only</td>
<td>Sputum +</td>
<td>2004</td>
<td>Susceptible</td>
<td>Susceptible</td>
<td>70337740001771</td>
<td>“Kilimanjaro”</td>
<td>227425113434</td>
</tr>
<tr>
<td><strong>Reactivations (indistinguishable strains)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5a Australia</td>
<td>Unknown</td>
<td>22</td>
<td>Lung plus pleura, Lung plus pleura</td>
<td>Sputum -, pleural fluid –, Sputum -</td>
<td>1995</td>
<td>Susceptible</td>
<td>Susceptible</td>
<td>777777777760771</td>
<td>T1</td>
<td>224325153223</td>
</tr>
<tr>
<td>5b Australia</td>
<td>Unknown</td>
<td>22</td>
<td>Lung plus pleura, Lung plus pleura</td>
<td>Sputum -</td>
<td>1997</td>
<td>Susceptible</td>
<td>Susceptible</td>
<td>777777777760771</td>
<td>T1</td>
<td>224325153223</td>
</tr>
<tr>
<td>6a Philippines</td>
<td>Unknown</td>
<td>11</td>
<td>Lung plus pleura, Lung plus pleura, genitourinary tract, other organ</td>
<td>Bronchial washings+, Sputum +, fine needle aspiration+</td>
<td>1996</td>
<td>Susceptible</td>
<td>Susceptible</td>
<td>677777477413671</td>
<td>EA12/“Manila”</td>
<td>264326223432</td>
</tr>
<tr>
<td>6b Philippines</td>
<td>Unknown</td>
<td>11</td>
<td>Lung plus pleura, Lung plus pleura, genitourinary tract, other organ</td>
<td>Sputum +</td>
<td>1998</td>
<td>Susceptible</td>
<td>Susceptible</td>
<td>677777477413671</td>
<td>EA12/“Manila”</td>
<td>264326223432</td>
</tr>
<tr>
<td>7a India</td>
<td>Unknown</td>
<td>21.5</td>
<td>Lung only, Lung only</td>
<td>Sputum +</td>
<td>1999</td>
<td>Susceptible</td>
<td>Susceptible</td>
<td>703777740003771</td>
<td>CAS1/“Delhi”</td>
<td>226415173333</td>
</tr>
<tr>
<td></td>
<td>Country</td>
<td>Status</td>
<td>Sequence</td>
<td>Sample Type</td>
<td>Year1</td>
<td>Year2</td>
<td>Susceptibility</td>
<td>Reference Number</td>
<td>City</td>
<td></td>
</tr>
<tr>
<td>---</td>
<td>---------</td>
<td>----------</td>
<td>----------</td>
<td>-------------------</td>
<td>-------</td>
<td>-------</td>
<td>----------------</td>
<td>------------------</td>
<td>--------------</td>
<td>---</td>
</tr>
<tr>
<td>7b</td>
<td></td>
<td></td>
<td></td>
<td>Pleura only</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>8a</td>
<td>Korea</td>
<td>Unknown</td>
<td>23</td>
<td>Lung only</td>
<td>2001</td>
<td></td>
<td>Susceptible</td>
<td>703777740003771</td>
<td></td>
<td></td>
</tr>
<tr>
<td>8b</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>9a</td>
<td>Australia</td>
<td>Negative</td>
<td>44.5</td>
<td>Lung only</td>
<td>2001</td>
<td></td>
<td>Susceptible</td>
<td></td>
<td>T1</td>
<td></td>
</tr>
<tr>
<td>9b</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10a</td>
<td>Thailand</td>
<td>Positive</td>
<td>1.5</td>
<td>Lung only</td>
<td>2005</td>
<td></td>
<td>MDR</td>
<td></td>
<td>EA15</td>
<td></td>
</tr>
<tr>
<td>10b</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>11a</td>
<td>Afghanistan</td>
<td>Negative</td>
<td>100</td>
<td>Lung only</td>
<td>1997</td>
<td></td>
<td>Susceptible</td>
<td>703777740003771</td>
<td>CAS</td>
<td></td>
</tr>
<tr>
<td>11b</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>2006</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>12a</td>
<td>Vietnam</td>
<td>Unknown</td>
<td>10</td>
<td>Lung only</td>
<td>1995</td>
<td></td>
<td>INH-R</td>
<td>N/A²²</td>
<td>N/A²²</td>
<td></td>
</tr>
<tr>
<td>12b</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1997</td>
<td></td>
<td>MDR</td>
<td></td>
<td>N/A²²</td>
<td></td>
</tr>
<tr>
<td>13a</td>
<td>Romania</td>
<td>Negative</td>
<td>9</td>
<td>Lung only</td>
<td>1995</td>
<td></td>
<td>Susceptible</td>
<td>N/A²²</td>
<td>N/A²²</td>
<td></td>
</tr>
<tr>
<td>13b</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1997</td>
<td></td>
<td></td>
<td></td>
<td>N/A²²</td>
<td></td>
</tr>
<tr>
<td>14a</td>
<td>Korea</td>
<td>Unknown</td>
<td>43</td>
<td>Lung only</td>
<td>1995</td>
<td></td>
<td>Susceptible</td>
<td>000000000003771</td>
<td>Beijing</td>
<td></td>
</tr>
<tr>
<td>14b</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1999</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>15a</td>
<td>United Kingdom</td>
<td>Unknown</td>
<td>5.5</td>
<td>Lung only</td>
<td>1999</td>
<td></td>
<td>Susceptible</td>
<td>77773777760771</td>
<td>T1</td>
<td></td>
</tr>
<tr>
<td>15b</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>2000</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
* Case number; (a) first disease episode (b) second disease episode

† Time period from cessation of treatment for first disease episode to recurrence of disease

‡ Year of isolation of strains

§ Susceptible means fully susceptible to all first-line antituberculous drugs; MDR = multi drug resistant (resistant to at least isoniazid and rifampicin); INH-R – resistant to isoniazid only

** Classification of spoligotypes as described by Bruday K et al. [20]

†† MIRU – mycobacterial interspersed repetitive unit typing as described by Supply et al. [16]

‡‡ By fine-needle aspiration.

§§ These pair of isolates were tested by IS6110 restriction fragment length polymorphism (RFLP) typing and found to be indistinguishable. The isolates are no longer available for testing by spoligotyping and MIRU typing
REFERENCES


mining the fourth international spoligotyping database (SpolDB4) for classification, population genetics and epidemiology. *BMC Microbiol* 2006; 6: 23.


Figure 1 Flow chart TB notifications 1994-2006

5723
Total TB notifications

8
Anonymous or incomplete name (no matching possible)

10
Duplicate notifications for same episode

5705
Identified notifications

1974
Culture negative single episodes and episodes with no information on cultures

3731
Culture positive single episodes

15
Culture positive recurrences after completed treatment

Figure 2 Kaplan-Meier curve showing risk of recurrence in 3731 culture positive tuberculosis notifications