# Tuberculosis in Australia: bacteriologically confirmed cases and drug resistance, 1998-1999

## *Report of the Australian Mycobacterium Reference Laboratory Network*

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## Abstract

**The Australian Mycobacterium Reference Laboratory Network collected and analysed laboratory data on new diagnoses of infection with *Mycobacterium tuberculosis* complex in 1998 and 1999. Totals of 700 and 760 cases were identified, representing annual reporting rates of 3.7 and 4.0 cases of laboratory confirmed tuberculosis (TB) per 100,000 population in the years 1998 and 1999 respectively. Australia's TB reporting rates have varied little in the past decade, ranging from 3.7 to 4.1 cases per 100,000 population. Reporting rates vary between States, reflecting differences in the distribution of persons in 'high-risk' categories for TB. The male:female ratio decreased to almost 1:1. The median age for males with culture-confirmed TB is in the 45-49 age group; for females, the median is in the 35-39 age group. Pulmonary disease was diagnosed in 63 per cent of cases whereas disease of lymph nodes accounted for 21 per cent of all cases. Children have the lowest rates of culture-confirmed TB; males in the older age groups have the highest rates. Microscopy was positive for 60 per cent of culture-positive sputa, and for approximately 45 per cent of bronchoscopy specimens. The frequency of multi-drug resistance (less than 1%) was slightly lower than in previous years. *Commun Dis Intell* 2001;25:261-265.**

*Keywords:* Mycobacterium *tuberculosis complex, laboratory network, tuberculosis, TB, drug resistance*

## *Introduction*

Data from the World Health Organization (WHO) show that Australia's notification rate for tuberculosis (TB) — around 5 cases per 100,000 population — is among the lowest in the world.[1] There remains, however, an undeniable need for surveillance among Australia's population in order to ensure the continued efficacy of our national TB program. Furthermore, reports from WHO point to the huge TB burden in certain neighbouring countries: they reflect the difficulties in managing TB in the presence of high background rates of tuberculous infection when health care systems are fragmented and under-funded.[2] Drug resistance and co-infection with HIV are additional obstacles to TB control in developing countries.[3]

Data pertaining to TB in Australia come from 2 sources. Since 1991, the National Mycobacterial Surveillance System (NMSS) of the Communicable Diseases Network Australia New Zealand has provided statistics on cases reported to public health authorities in Australia's States and Territories.[4] The Australian Tuberculosis Reporting Scheme has been conducted by the Mycobacterium Reference Laboratory Network (MRLN) since 1986.[5] Whereas a proportion of cases registered with NMSS will have been identified through clinical and epidemiological criteria alone, the statistics compiled by the MRLN relate to diagnoses made by isolation of *Mycobacterium tuberculosis* complex. This report deals with laboratory diagnoses made in the years 1998 and 1999.

## *Methods*

The data are based on patients submitting clinical samples from which *Mycobacterium tuberculosis* complex (MTBC, but excluding the BCG strain) are grown on culture. Due to the specialised nature of TB bacteriology, it can be assumed that the 5 laboratories that comprise the MRLN account for almost all, if not all, of the bacteriological diagnoses in Australia. Comparable bacteriological procedures are used in the reference laboratories. Relapse patients, that is, those previously diagnosed, treated and considered cured, were included in these data because laboratories cannot usually differentiate them from new cases. Temporary visitors to Australia are also included.

For each new laboratory diagnosis the following information was collected:

- demographic: patient identifier, age, sex, HIV status and state of residence
- specimen: type, site of collection, date of collection and microscopy result, and
- isolate: species of mycobacterium and results of drug susceptibility tests.

Data for 1998 and 1999 from contributing laboratories were submitted in standard format to the scheme co-ordinator for collation and analysis. Duplicate entries (as indicated by identical patient identifier and age) were deleted before analysis. Rates were calculated using the respective mid-year estimates of the population supplied by the Australian Bureau of Statistics.

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The nature of the first clinical sample that yielded an isolate of MTBC was used to record the nominal site of disease for individual cases. Culture-positive specimens collected at bronchoscopy, as well as gastric washings, were taken to identify cases of pulmonary disease. In cases of multi-site disease, provided a sputum sample was culture-positive, these cases were included among those listed as having pulmonary disease: the most significant category for public health purposes. Although many patients were known to have isolates from more than one body site, such data are of doubtful value for the laboratory-based report and were not collated. Similarly, it is not always possible to accurately categorise cases of miliary and disseminated disease from data available to laboratories.

## *Results*

### Total reports and distribution by State

Totals of 700 and 760 cases were recorded in 1998 and 1999 respectively. These figures represent annual rates of 3.7 and 4.0 cases of laboratory-confirmed TB per 100,000 population. The distribution of cases by state of residence is shown in Table 1. State-specific annual reporting rates varied from less than one (Tasmania, 1999) to 11.6 per 100,000 (Northern Territory, 1998). It should be noted that the data from Victoria includes 37 cases identified among Timorese refugees.

### Causative organism

The large majority of cases were due to *M. tuberculosis*. However, in 1998 there were 5 isolates of *M. bovis* and two of *M. africanum*. In 1999, there were 4 isolates of *M. bovis* and three of *M. africanum*.

### Distribution by gender, age and site of disease

Full information for gender, age and site of disease was submitted for 688 of the 700 cases recorded in 1998, and for 756 of the 760 cases recorded in 1999. Figure 1 shows the distribution of the 1444 cases by age group and gender. The overall male:female ratio was 0.96:1 in 1998 and 1.15:1 in 1999. For the 2 years combined it was 1.06:1. In both years, the median age group for all cases was 40-44 years. The median age group for males was 45-49 whereas that for females was 40-44 years. Age and gender specific rates varied from less than one per 100,000 population in children younger than 15 years to almost 30 per 100,000 per year in males over 80 years of age (data not shown). Nine cases were children younger than 10 years. Four children had disease in pulmonary sites, four had lymph node infections, and one had disease in a knee joint. There were no culture-confirmed cases of tuberculous meningitis in children. (**Editor's note:** There were 13 cases of tuberculous meningitis reported in the preceding TB Annual report, 1999 (pp 254-259). All but 2 of these were in adults. Of the two cases in children, one was culture-confirmed and one was diagnosed by microscopy.)

Figure 2 shows the distribution of 1,444 cases by site of disease and sex. Pulmonary infection was demonstrated in 63 per cent of the total cases (male:female ratio 1.3:1). Several patients with pulmonary disease were also proven to have disease in other body sites. Twenty-one per cent had disease of lymph nodes identified (male:female ratio 0.5:1). There were also 60 cases of pleural disease (male:female ratio 1.6:1) and 50 cases of disease in genito-urinary sites (male:female ratio 1.0:1).

### Association with HIV

The reference laboratories reported no isolates of MTBC from persons known to be HIV+. For the majority of patients, however, HIV status was not recorded.

**Figure 1. MTBC isolates, 1998 to 1999, by age group and sex**

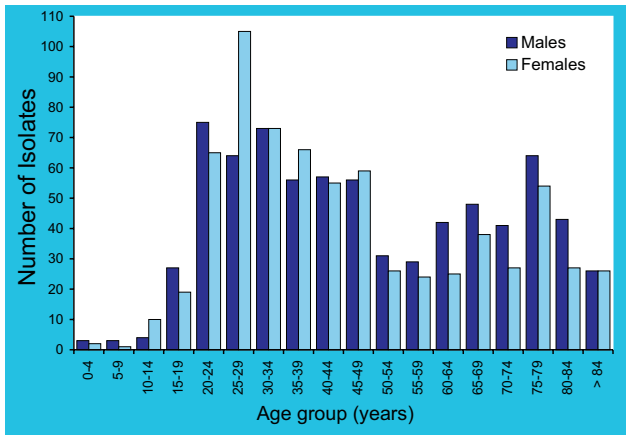
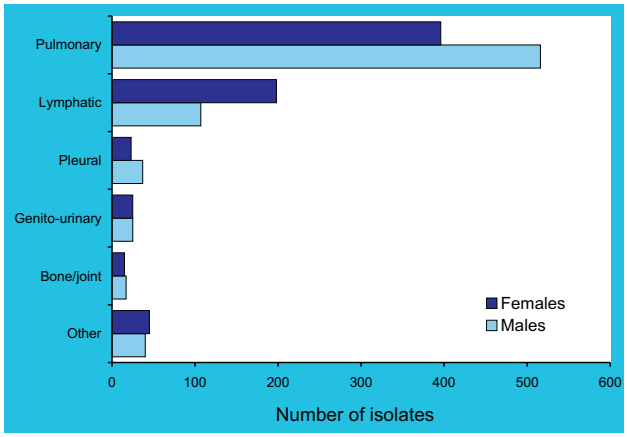



**Table 1. MTBC isolates in Australia, 1996 to 1999, cases and rates per 100,000 population, by State or Territory**

| State | 1999 | | 1998 | | 1997[1] | | 1996[1] | |
|---|---|---|---|---|---|---|---|---|
| | n | Rate | n | Rate | n | Rate | n | Rate |
| New South Wales[2] | 291 | 4.3 | 289 | 4.4 | 329 | 5.0 | 341 | 5.3 |
| Victoria | 261 | 5.5 | 192 | 4.1 | 193 | 4.2 | 214 | 4.7 |
| Queensland | 75 | 2.1 | 85 | 2.5 | 74 | 2.2 | 90 | 2.7 |
| Western Australia | 64 | 3.4 | 66 | 3.6 | 51 | 2.8 | 51 | 2.9 |
| South Australia | 46 | 3.1 | 40 | 2.7 | 39 | 2.6 | 28 | 1.9 |
| Tasmania | 2 | 0.4 | 6 | 1.3 | 8 | 1.8 | 3 | 0.6 |
| Northern Territory | 21 | 10.9 | 22 | 11.6 | 28 | 15.0 | 23 | 12.6 |
| Total | 760 | 4.0 | 700 | 3.7 | 722 | 3.9 | 750 | 4.1 |

1. Data from previous reports of the MRLN.
2. Data for the Australian Capital Territory are included with those from New South Wales.

**Figure 2. MTBC isolates, Australia, 1998 to 1999, by site and sex**




## *Microscopy results*

Acid-fast microscopy (AFM) results were available for 677 and 753 samples in 1998 and 1999 respectively (Table 2). In 1998, 60.3 per cent of 300 sputum samples that were positive on culture were also positive by AFM; the corresponding figures for 1999 were 60.9 per cent of 396 sputum samples. Bronchoscopy samples provided 87 diagnoses in 1998 in which AFM was positive for 41 (47.1%) infections. In 1999, 40 (43.5%) of 92 bronchoscopy collections were positive on smear.

### *In vitro* drug susceptibility

In 1998, 699 of 700 isolates, and in 1999 all 760 isolates, were tested for *in vitro* susceptibility to the 4 drugs recommended for standard treatment of TB in Australia, i.e., isoniazid (H), rifampicin (R), ethambutol (E) and pyrazinamide (Z).[6] A total of 68 isolates (9.7% of the total) in 1998 were resistant to at least one of the standard compounds; the corresponding figure for 1999 was 59 (7.8%). The frequency of resistance to H, R, E and Z, alone or in combination, is shown in Table 3. Resistance to H and/or R was recorded in 65 isolates (9.2% of total) in 1998 and in 54 isolates (7.1%) in 1999. Resistance to both H and R ('multi-drug resistant TB', MDR-TB) was demonstrated in 6 isolates (0.9% of total) in 1998 and 4 (0.5%) isolates in 1999 (Table 4). All of the MDR isolates were *M. tuberculosis*. Eight of the 10 MDR isolates came from pulmonary sites; two were from cervical lymph nodes. Five patients were diagnosed from sputum samples, of which three were

**Table 2. Frequency of positive microscopy in specimens yielding MTBC on culture in 1998 and 1999**

| | 1999 | | | 1998 | | |
|---|---|---|---|---|---|---|
| | No | Smear positive | % | No | Smear positive | % |
| All specimens | 753 | 368 | 48.9 | 677 | 303 | 44.7 |
| Sputum | 396 | 241 | 60.9 | 300 | 181 | 60.3 |
| Bronchoscopy | 92 | 40 | 43.5 | 87 | 41 | 47.1 |

**Table 3. *In vitro* resistance of isolates to the standard anti-tuberculosis drugs, Australia, 1996 to 1999**

| | 1999 | | 1998 | | 1997[1] | | 1996 | |
|---|---|---|---|---|---|---|---|---|
| | No | %[2] | No | %[2] | No | %[2] | No | %[2] |
| Isoniazid (H) | 52 | 6.8 | 63 | 9.9 | 48 | 6.6 | 73 | 9.7 |
| Rifampicin (R) | 6 | 0.8 | 8 | 1.1 | 15 | 2.1 | 16 | 2.1 |
| Ethambutol (E) | 2 | 0.3 | 4 | 0.6 | 4 | 0.6 | 2 | 0.3 |
| Pyrazinamide[3] (Z) | 6 | 0.8 | 9 | 1.3 | 24 | 3.3 | 18 | 2.3 |

1. Data taken from previous publications of the MRLN.
2. Percentage of strains tested which were resistant to drug alone or in combination with other drugs
3. All strains of *M. bovis* are naturally resistant to pyrazinamide; 5 *M. bovis* were identified in 1998; 4 *M. bovis* were identified in 1999.

**Table 4. Drug resistance patterns in MDR strains, Australia, 1996 to 1999**

| Resistance pattern (standard drugs)[1] | No of isolates | | | |
|---|---|---|---|---|
| | 1999 | 1998 | 1997 | 1996 |
| H + R only | 2 | 2 | 6 | 10 |
| H + R + E | 1 | 1 | 1 | 1 |
| H + R + Z | 1 | 2 | 5 | 4 |
| H + R + E + Z | 0 | 1 | 2 | 0 |

1. H = isoniazid; R = rifampicin; E = ethambutol; Z = pyrazinamide

positive by AFM. Six of 37 isolates from Timorese patients were resistant to H; none were MDR. Five isolates identified as *M tuberculosis* were recorded as resistant to Z alone. In addition to the standard drugs, streptomycin (S) was tested against the majority of isolates showing resistance to H and/or R as well as 348 isolates that were fully-susceptible to the standard drugs. Approximately 30 per cent of strains resistant to H and/or R are also resistant to S and a further 4 per cent of otherwise susceptible strains are resistant to S.

## *Discussion*

The data for 1998-99 confirm the continuing stable situation for bacteriologically-confirmed TB in Australia: annual reporting rates of approximately 4 cases per 100,000 per year have been recorded for the past 10 years. The pathophysiology of TB dictates that not all cases reported from clinical sources will be confirmed by laboratories. This situation will apply even though an appropriate clinical sample was submitted to a competent laboratory for microbiological testing. Data from NMSS showed that 923 cases of tuberculosis were notified by State and Territory jurisdictions during 1998; reference laboratories confirmed only 700 cases in that year. This finding is in keeping with previous comparative data,[7] and suggest that in Australia, at present, only around 75 per cent of cases reported to NMSS are confirmed by culture. The Public Health Laboratory Network's definition of a laboratory-proven case of TB allows for detection of specific nucleic acid in clinical samples, in the absence of confirmation by a positive culture. Wider use of nucleic acid amplification assays, and a better appreciation of the clinical utility of such tests, may bring a reduction in the numerical disparity between clinical and laboratory diagnoses. Future reports from the MRLN will include diagnoses made by nucleic acid amplification tests.

The annual incidence rates in the various States and Territories range from below 2 per 100,000 in Tasmania to more than 11 per 100,000 in the Northern Territory (Table 1). Current data show only minor deviations from those in previous years. As would be expected, when compared to laboratory-derived data, the NMSS reports provide relatively similar state-specific differences in case numbers and reporting rates — which can be taken as validation of the parallel reporting processes. The differences in rates between States and Territories are almost certainly due to peculiarities in the demography of high-risk subgroups, rather than local differences in the risk of acquiring tuberculous infection.

Our previous report made mention of incomplete data relating to age, sex and site of disease.[7] The ascertainment level for these data is much improved; 688 of 700 cases in 1998, and 756 of 760 cases in 1999. Cases of active disease are distributed unevenly across age groups (Figure 1). The distributions shown in Figure 1 are almost identical to those in previous reports, and generally agree with statistics from notified cases. NMSS data show significantly more cases in young children, e.g. 28 reports in children under 10 years were notified to NMSS, whereas only 3 cases were confirmed by culture. This discrepancy can be explained by the fact that diagnosis of TB in young children is difficult, and is often based on clinical and epidemiological findings, rather than definitive laboratory results.[8]

There are signs that the overall male:female ratio – which has previously been reported as around 1.3:1 – is becoming closer to 1:1. For the current reporting period, there were almost equivalent numbers of males and females (1.06:1). NMSS data for 1988 generally agree with this observation. Our laboratory data show clear differences in sex-based distribution within the various disease categories. Pulmonary and pleural TB is more likely to be found in males, whereas the distribution of lymphatic TB is heavily skewed to females (male:female ratio, 0.5:1). Earlier studies by the MRLN have consistently found more cases of lymph node disease in females.

Pulmonary infection was demonstrated in 63 per cent of patients with culture-proven TB. The majority of patients who did not have pulmonary TB had disease of the lymph nodes (21 per cent of the total). In 1996-97 corresponding figures were 64 per cent and 19 per cent. The NMSS report for 1998 gives figures of 59.3 and 21.5 per cent for pulmonary and lymph node infection, respectively.[4]

Our data illustrate the continuing value of microscopy as a diagnostic tool (Table 2). Almost half of all samples that grew MTBC on culture were found positive on smear. Among patients with pulmonary disease, 60 per cent of the culture-positive sputum specimens were also positive by AFM. United States authorities report an equivalent statistic for AFM in that country.[9] We found that microscopy was less sensitive on bronchoscopy samples, detecting around 45 per cent of culture-positive samples. The 1998 report from NMSS indicated that only 32 per cent of pulmonary cases were smear-positive.[4] While this might suggest under-reporting of microscopy results in NMSS, it should be noted that the denominator in the NMSS statistic would include:

(i) diagnoses made from histology, and

(ii) diagnoses made on clinical findings alone.

The median age group for females with TB (40-44) is higher than in 1997 when it was 35-39. The median age group for males is unchanged at 45-49 years. NMSS data has shown different age distributions of Australian-born patients compared to counterparts born overseas, with the former generally being older than the latter.[4] Persons born outside Australia now account for three-quarters of Australian TB notifications and provide the majority of cases in the young and middle age groups.

Although there were no culture-proven cases listed as associated with HIV, this is almost certainly not the true picture. Reference laboratory databases are unlikely to show HIV status for patients under investigation for TB. NMSS reported 4 cases of HIV associated TB in 1998.[4]

This report provides recent data on *in vitro* drug resistance among isolates from Australian patients with TB. To date, such information is not available through NMSS. We found that 9.7 per cent and 7.8 per cent of isolates during 1998 and 1999 respectively, had in vitro resistance to at least one of the standard anti-TB drugs, H, R, E or Z. The corresponding figures for previous years were: 1997 (9%); 1996 (11%); 1995 (9%); 1994 (7%). Resistance to one or both of H and R — the most effective anti-tuberculosis compounds — was detected in 9.2 per cent and 7.1 per cent of isolates in 1998 and 1999 respectively. Corresponding figures for previous years were: 1997 (6.9%); 1996 (9.9%); 1995 (7.5%); 1994 (6.1%). We found 6 (0.9%) isolates in 1998 and 4 (0.5%) in 1999 that were resistant to H and R, i.e., were MDR. The

corresponding figures for previous years were: 1997 (1.9%); 1996 (2%); 1995 (0.7%); 1994 (0.3%). Our findings thus show no significant temporal changes in the prevalence of drug resistance among Australian isolates.

Laboratories do not have the information necessary to stratify patients on the basis of prior anti-tuberculosis therapy. It is therefore not possible to categorise resistance as one of 'primary' or 'acquired'. The supplementary dataset now established for cases notified to NMSS has the potential to allow more precise correlation of drug resistance with a patient's country of birth and treatment history. Such data are required by the Global Project on Anti-tuberculosis Drug Resistance Surveillance. Anecdotal evidence suggests that acquired drug resistance is rarely seen in patients receiving treatment in Australia. Furthermore, drug resistance is almost always linked to persons born outside Australia, or to the Australian-born who acquire tuberculous infection when travelling overseas (Dawson, unpublished). This observation, combined with the demonstrated low prevalence of drug resistance, justifies the conclusion that transmission of a drug-resistant strain of MTBC is an uncommon event in this country. Outbreaks of drug-resistant TB are unquestionably rare in Australia.

Molecular studies of resistant isolates of MTBC have defined the specific mutations that confer their resistance to the anti-tuberculosis drugs.[10] These studies have provided insight into the mechanisms of drug-resistance and through correlation with MIC values have confirmed earlier reports of strains with 'low-level' and 'high-level' resistance to H.[11] A significant proportion of H-resistant isolates have MIC values of 0.1 mcg/ml, while others are resistant at 0.4 mcg/ml. The clinical implication of these findings have not been proven, although the argument that 'low-level' resistance is likely to respond to standard therapy with H, would seem tenable. All Australian reference laboratories are now testing for resistance at both levels of H. Future reports will attempt to provide more detailed data on this topic.

## *Acknowledgements*

The Australian Mycobacterium Reference Laboratory Network comprises the Mycobacterium Reference Laboratories at the following institutes:

Queensland Health Pathology Services, The Prince Charles Hospital, Chermside, Queensland.

Institute of Clinical Pathology and Medical Research, Westmead Hospital, Westmead, New South Wales.

Victorian Infectious Diseases Reference Laboratory, North Melbourne, Victoria.

Institute of Medical and Veterinary Sciences, Adelaide, South Australia.

Western Australian Centre for Pathology and Medical Research, The Queen Elizabeth II Medical Centre, Nedlands, Western Australia.

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