

Continuing *Mycobacterium bovis* transmission from animals to humans in New Zealand

M. G. BAKER^{1*}, L. D. LOPEZ¹, M. C. CANNON², G. W. DE LISLE²
AND D. M. COLLINS²

¹ Institute of Environmental Science and Research Ltd (ESR), Porirua, New Zealand

² AgResearch, Wallaceville, Upper Hutt, New Zealand

(Accepted 10 December 2005, first published online 29 March 2006)

SUMMARY

New Zealand has a large reservoir of *Mycobacterium bovis* infection in wild and farmed animals. This study aimed to assess the extent of human infection with this organism and the potential contribution of these animal sources. Combined epidemiological and laboratory investigation of human tuberculosis cases over the period 1995–2002 showed that *M. bovis* accounted for 2·7% (54/1997) of laboratory-confirmed human tuberculosis cases, a rate of 0·2/100 000 population. *M. bovis* isolates from humans (23) were typed using restriction endonuclease analysis (REA) and compared with isolates from wild and domestic animals (2600). Fourteen (61%) of the human isolates had REA patterns that were identical to patterns for isolates from cattle, deer, possums, ferrets, pigs, and occasionally cats. These results suggest a low level of ongoing *M. bovis* transmission from animal reservoirs to humans in New Zealand.

INTRODUCTION

Mycobacterium bovis can cause disease in humans that clinically is indistinguishable from that due to *Mycobacterium tuberculosis*. Although now uncommon in most developed countries, *M. bovis* remains an important cause of tuberculosis in developing countries [1, 2]. *M. bovis* is a common infection in wild animals in New Zealand, notably introduced Australian brushtail possums (*Trichosurus vulpecula*), and these occasionally infect farmed cattle and deer [3]. This situation raises the potential for animal-to-human transmission of this pathogen.

A previous review of human infection with *M. bovis* in New Zealand, identified 60 cases over the period from January 1985 to July 1995 [4]. The majority of

these cases occurred in elderly people and probably represent infections acquired before introduction of milk pasteurization and herd testing in the 1940s and 1950s. However, this total included some younger cases (15 under 40 years of age) who should not have been exposed to unpasteurized milk.

This study, therefore, aimed to describe the current epidemiology of human *M. bovis* infection in New Zealand and determine the possible sources of these infections using combined epidemiological and laboratory investigation of cases over the 1995–2002 period.

METHODS

Human *M. bovis* and *M. tuberculosis* infections from 1995 to 2002 were identified using notification records supplemented by laboratory detection data from New Zealand's three tuberculosis referral laboratories. Cases occurring from 1998 to 2002 were analysed in

* Author for correspondence: Dr M. G. Baker, Department of Public Health, Wellington School of Medicine & Health Sciences, Box 7343, Wellington South, New Zealand.
(Email: michael.baker@wnmeds.ac.nz)

detail, including key demographic and risk factor information. This period was chosen because few human isolates were stored before 1998. Rates were calculated using population data from the 2001 Census. Epi-Info (CDC, Atlanta, GA, USA) was used to calculate crude rate ratios (RR) and 95% confidence intervals 95% CI and the χ^2 test for linear trend.

The three tuberculosis laboratories sent all stored human *M. bovis* isolates to AgResearch, Wallaceville for DNA fingerprinting. AgResearch carried out this characterization of human isolates using restriction endonuclease analysis (REA). Each isolate was separately digested with three restriction enzymes, *BstEII*, *PvuII* and *BclI*. This process generated three gels and associated band pattern for each isolate. This was the same method as had been used to type 2600 *M. bovis* isolates collected from a wide variety of domestic and wild animals in New Zealand over the 1982–2003 period [5–7]. These animal isolates had been characterized into 270 different REA types and the information stored in a DNA database. The restriction types of the human *M. bovis* isolates were compared to these animal types.

The epidemiological information was analysed in conjunction with the results of the DNA fingerprinting of the *M. bovis* strains recovered from the patients. This process aimed to determine the likelihood of the human infections being acquired from specific animal exposures in New Zealand or from overseas.

RESULTS

Epidemiology of *M. bovis* infection

Notification and laboratory data identified a total of 54 human cases of *M. bovis* (excluding *M. bovis* BCG) infection from 1995 to 2002. There were a further 1943 cases of infection with *M. tuberculosis*, so *M. bovis* represented 2.7% (54/1997) of the laboratory-confirmed tuberculosis cases detected over that period.

The incidence of newly diagnosed *M. bovis* infections (Fig. 1) did not increase over the period 1995–2002 (χ^2 for linear trend, $P=1.0$). The average number of bovine tuberculosis cases in the 8-year period was 6.8 per year.

The 34 *M. bovis* infections occurring from 1998 to 2002 were reviewed in detail. Over this 5-year period, cases occurred in 16 of New Zealand's 21 district health board areas, with an average rate of 0.18/100 000 cases. Rates ranged from 0 to 1.32/100 000,

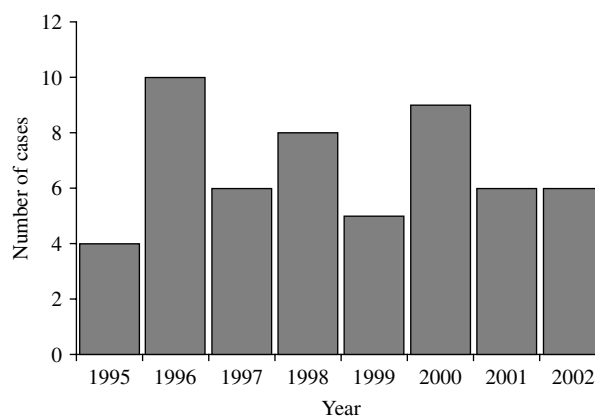


Fig. 1. Human *M. bovis* infections in New Zealand by year, 1995–2002.

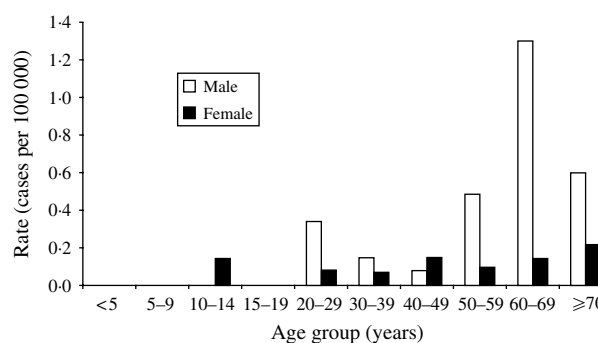


Fig. 2. Human *M. bovis* infection incidence by age and sex, average annual rate per 100 000 population, 1998–2002.

although these figures are based on small numbers. Areas reporting rates above the national average were West Coast, Wairarapa, Hutt, Lakes, Tairāwhiti, Waikato, Counties Manukau, Eastern Bay of Plenty, Capital Coast and Whanganui districts.

Figure 2 shows the average annual rates of *M. bovis* infection by age group and sex. Highest rates were in the 60–69 years age group, followed by those ≥ 70 years of age. Gender was recorded for all 34 cases. Male cases outnumbered females by 3:1.

Country of birth information was available for 32 cases of whom 23 (71.9%) were born in New Zealand and the remaining nine (28.1%) had migrated to New Zealand. Six of the immigrant cases had been born in developing countries. The rates for Maori (2.1/100 000) and Pacific people (1.5/100 000) and people of other ethnicity (1.2/100 000) were higher than for Europeans (0.5/100 000).

The site of infection was recorded for 88.2% of cases (30/34). Pulmonary infection was the most common site, accounting for 56.7% (17/30). The remaining extra-pulmonary sites included: peritoneum,

Table 1. Comparison of the epidemiological characteristics of human *M. bovis* and *M. tuberculosis* infections, 1998–2002

Characteristic	<i>M. bovis</i> infection (% recorded)	<i>M. tuberculosis</i> infection (% recorded)
Sex		
Male	25 (73.5)	696 (52.4)
Female	9 (26.5)	632 (47.6)
Median age (yr)	57	37
Age group (yr)		
0–19	1 (2.9)	153 (11.5)
20–59	17 (50.0)	866 (64.9)
≥60	16 (47.1)	316 (23.7)
Ethnicity		
European	14 (45.2)	169 (13.1)
Maori	11 (35.5)	217 (16.8)
Pacific	3 (9.7)	240 (18.6)
Other	3 (9.7)	665 (51.5)
Region		
North Island	26 (76.5)	1183 (88.5)
South Island	8 (23.5)	153 (11.5)
Site		
Pulmonary	17 (56.7)	685 (56.9)
Non-pulmonary	10 (33.3)	383 (31.8)
Both pulmonary and non-pulmonary	3 (10.0)	134 (11.1)
Birth		
Inside New Zealand	23 (71.9)	360 (29.6)
Outside New Zealand	9 (28.1)	855 (70.4)
Total 1998–2002	34	1336

lymph nodes, arm/wrist (tenosynovitis), genitourinary system, hip joint, and proximal interphalangeal joint. Most cases were hospitalized (19/26 or 73.1%). The case-fatality rate was 15.4% (4/26).

Comparison of *M. bovis* and *M. tuberculosis* infection

Table 1 compares the characteristics of *M. bovis* and *M. tuberculosis* cases for the 5-year period 1998–2002. People infected with *M. bovis* were significantly more likely to be male (RR 2.5, 95% CI 1.2–5.3), over 60 years of age (RR 2.5, 95% CI 1.2–5.3), European (RR 6.2, 95% CI 3.1–12.1) or Maori (RR 2.2, 95% CI 1.1–4.6), to have been born inside New Zealand (RR 4.6, 95% CI 2.0–10.6) rather than migrated there, and to be living in the South Island at the time of diagnosis (RR 2.3, 95% CI 1.1–5.0). Of note is that *M. bovis* infection was not more commonly associated with extra-pulmonary sites of infection than was *M. tuberculosis*.

Laboratory investigation of human *M. bovis* infections

The three tuberculosis referral laboratories located 23 viable human *M. bovis* isolates for the 1998 to 2002 period, out of a possible total of 34. The AgResearch laboratory at Wallaceville typed these isolates and compared their patterns to REA results for animal isolates held in their database (Table 2).

Eight isolates were classified as ‘foreign’ strains as their REA pattern has not been seen previously in a New Zealand animal isolate. Of this group, four of the source patients were recorded as having been born overseas (one in Fiji, one in the Cook Islands, one in England and one in Scotland), and four were born in New Zealand. One isolate was the strain (AN5) used for making bovine purified protein derivative (PPD). This infection was a result of an accidental exposure that occurred in a laboratory manufacturing tuberculin.

This left 14 isolates that had an REA pattern that was identical to one that has been seen previously in wild or domestic animals in New Zealand. These isolates included nine different unique REA patterns. Of these, five isolates had the same REA type (115) that is found very commonly in the Central North Island in a wide range of animal reservoirs. Two isolates had the same type (21) that is found very commonly in the Wairarapa and on the West Coast of the South Island, again in a wide range of reservoir animals. The remaining seven REA types were all represented by a single isolate. The types found in humans had all been isolated from bovine reservoirs (14/14 isolates). There was also a high degree of overlap with those found in deer (12/14), ferrets (10/14) and possums (11/14).

Of these 14 cases with REA patterns that matched specimens from animals in New Zealand, eight had risk factor information recorded. Of these, five reported animal contact that could potentially have given rise to infection. These contacts included living or working on a farm, being an abattoir worker, consumption of unpasteurized milk, and working as a veterinary receptionist. One case was infected following accidental inoculation during necropsy of a tuberculous possum, as specifically reported elsewhere [8].

DISCUSSION

Findings from this study suggest a continuing low level of *M. bovis* transmission from animals to humans in New Zealand. Although *M. bovis* is a relatively uncommon cause of human tuberculosis in New

Table 2. Human *M. bovis* isolates (1998–2002), and animal isolates (1982–2003) by REA type (see references [5–7] for details of REA types)

REA type	No. of human isolates (% of total human isolates)	No. of animal isolates	No. of isolates from each animal species (% of total isolates for that species)					
			Bovine	Cervine	Ferret	Possum	Porcine	Feline
Foreign*	8 (34.8)	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
115	5 (21.7)	417	308 (24.1)	51 (10.6)	15 (7.7)	27 (6.4)	15 (8.1)	1 (2.4)
21	2 (8.7)	97	41 (3.2)	15 (3.1)	11 (5.6)	18 (4.3)	11 (5.9)	1 (2.4)
AN5*	1 (4.3)	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
6	1 (4.3)	3	3 (0.2)	0	0	0	0	0
9	1 (4.3)	3	2 (0.2)	1 (0.2)	0	0	0	0
19	1 (4.3)	33	7 (0.5)	24 (5.0)	1 (0.5)	1 (0.2)	0	0
27	1 (4.3)	30	1 (0.1)	3 (0.6)	0	13 (3.1)	13 (7.0)	0
62	1 (4.3)	109	50 (3.9)	11 (2.3)	16 (8.2)	10 (2.4)	17 (9.2)	5 (11.9)
151	1 (4.3)	111	78 (6.1)	18 (3.7)	7 (3.6)	6 (1.4)	1 (0.5)	1 (2.4)
158	1 (4.3)	3	3 (0.2)	0	0	0	0	0
Other	0	1794	783 (61.4)	359 (74.5)	146 (74.5)	344 (82.1)	128 (69.2)	34 (81.0)
Total	23	2600	1276	482	196	419	185	42

REA, Restriction endonuclease analysis; n.a., not applicable.

* Never found in animals in New Zealand.

Zealand, accounting for about 2.7% of human disease, this contribution is higher than that seen in other developed countries. In Australia *M. bovis* caused ~1% of human disease over the 1970–1994 period [9] and in Britain it accounted for 1.2% of cases from 1986 to 1990 [10]. Another indication of *M. bovis* transmission within New Zealand comes from the observation that most cases (71.9% over the 1998–2002 period) were born in New Zealand. This proportion is significantly higher than that seen for infection with *M. tuberculosis* where a minority of cases are New Zealand born (only 29.6% during this period).

There has been little increase over 17 years in the incidence of human infection with *M. bovis* in New Zealand. A previous analysis by Richard Pooley identified 60 cases over the 11½-year period from January 1985 to July 1995, an average of 5.2 cases a year [4]. This present study identified an average of 6.8 cases a year for the 8-year period from 1995 to 2002.

The site of tuberculosis infection gives an indication of the possible route of transmission. The majority (56.7%) of *M. bovis* infections are pulmonary, which is a similar proportion to that seen for *M. tuberculosis* infection. This contrasts with the pattern prior to pasteurization of milk in the 1940s when infection was frequently transmitted via contaminated milk and the majority of infections were non-pulmonary [11].

The ages of human *M. bovis* cases suggest that many are reactivations of old infections rather than

newly acquired disease. Among the New Zealand-born cases, most (80%) were >30 years of age which is similar to the 85% found by Pooley [4]. The median age of *M. bovis* infections (57 years) is much older than that for *M. tuberculosis* infection (37 years). Despite this, there are still *M. bovis* infections occurring in relatively young New Zealanders with four <30 years of age at the time of diagnosis. They were all born after control measures were introduced to limit transmission through contaminated milk. These measures included the compulsory testing of dairy cattle from 1961 and steady introduction of milk pasteurization from the 1940s to the early 1970s. These cases are likely to have acquired infection from other sources. The 3:1 preponderance of males compared to females suggests that occupational settings may be important sources of *M. bovis* exposure. A similar overrepresentation of males is seen for leptospirosis in New Zealand, and has been attributed to the larger proportion of males working as farm and abattoir workers where most exposure to this pathogen occurs [12].

Combined results of molecular and epidemiological investigation can provide evidence of the sources of human infections. For the two *M. bovis* infections resulting from well-documented occupational exposures, the source of infection was clearly identified. Five other cases in this present study reported animal contact that could potentially have given rise to

infection and had REA patterns that were the same as ones found in wild and domestic animals in New Zealand. Here the evidence is suggestive of local infection but more detailed investigation would be required to identify specific sources. Far less can be concluded about the source of infection for the remaining cases, including the eight others with REA patterns that were the same as ones found in wild and domestic animals in New Zealand.

There are few reported studies that have used molecular methods to investigate sources of human *M. bovis* infection. One study analysed 20 human and 58 animal isolates from Argentina and 47 human and 13 animal isolates from The Netherlands using restriction fragment length polymorphism (RFLP) patterns probed by the insertion element IS6110 [13]. It found that most human isolates in Argentina were of a single type that also predominated among cattle isolates. This situation contrasted with The Netherlands where the *M. bovis* fingerprints of strains from humans were very diverse and none were found among *M. bovis* strains of animal origin. A study in Sweden used similar methods to characterize isolates from 11 humans and 28 farmed deer sampled from an outbreak affecting five herds. The *M. bovis* fingerprints of the human isolates were diverse, whereas the isolates from the deer all had a different and very specific pattern [14]. The molecular epidemiology of human *M. bovis* infection in New Zealand, characterized by human infection with a diverse range of REA types, resembles the situation seen in The Netherlands and Sweden more than that in Argentina, but differs from Sweden and Argentina in having many of these types infect both animals and humans.

This study cannot exclude human sources for some *M. bovis* infections. For example, one case in our series reported contact with another case of human *M. bovis* infection. Human-to-human [15], and even human-to-animal transmission of *M. bovis* has been documented [16], and a recently reported contact investigation study suggests that in some circumstances *M. bovis* may be almost as efficient at human-to-human transmission as *M. tuberculosis* [17]. Despite these observations, humans are usually a 'dead-end' host for *M. bovis* infections and human contact is not generally an important source of infection.

The major limitations of this present study are its small size and the lack of complete epidemiological information on human cases. Continuing the typing of human isolates prospectively would increase the

size of this study sample over time. It would also be useful to undertake more detailed epidemiological investigation of these human cases, including their history of animal contact and geographic areas where they have lived in the past, so that these results could be compared with the extensive information held on *M. bovis* distribution in animals. A case-control study should also be considered to provide a more precise estimate of the risk of human infection associated with specific forms of animal contact.

ACKNOWLEDGEMENTS

Staff of New Zealand's three tuberculosis referral laboratories are acknowledged for the isolation of the *M. bovis* strains and forwarding them to Wallaceville for DNA fingerprinting: Ross Vaughan, Mycobacteria Laboratory, Auckland Hospital; Jane Wakefield, Kathryn Coley, Vivienne Talbot, Mycobacteria Laboratory, Waikato Hospital; Leo McKnight, Mycobacteria Laboratory, Wellington Hospital. Gary Yates, AgResearch, is thanked for culturing the *M. bovis* isolates for DNA fingerprinting. This study was contracted by the Animal Health Board.

DECLARATION OF INTEREST

None.

REFERENCES

1. **Grange JM.** *Mycobacterium bovis* infection in human beings. *Tuberculosis (Edinburgh)* 2001; **81**: 71–77.
2. **Ayele WY, et al.** Bovine tuberculosis: an old disease but a new threat to Africa. *International Journal of Tuberculosis and Lung Disease* 2004; **8**: 924–937.
3. **O'Neil BD, Pharo HJ.** The control of bovine tuberculosis in New Zealand. *New Zealand Veterinary Journal* 1995; **43**: 249–255.
4. **Pooley RB.** *Mycobacterium bovis*: an old disease in a new era? A review of the epidemiology and public health importance of human *Mycobacterium bovis* infection in New Zealand [Masters thesis]. Christchurch: University of Otago, 1996.
5. **Collins DM, de Lisle GW.** DNA restriction endonuclease analysis of *Mycobacterium bovis* and other members of the tuberculosis complex. *Journal of Clinical Microbiology* 1985; **21**: 562–564.
6. **Collins DM, de Lisle GW, Gabric DM.** Geographic distribution of restriction types of *Mycobacterium bovis* isolates from brush-tailed possums (*Trichosurus vulpecula*) in New Zealand. *Journal of Hygiene (London)* 1986; **96**: 431–438.

7. **Collins DM, Gabric DM, de Lisle GW.** Typing of *Mycobacterium bovis* isolates from cattle and other animals in the same locality. *New Zealand Veterinary Journal* 1988; **36**: 45–46.
8. **Cooke MM, et al.** Accidental *Mycobacterium bovis* infection in a veterinarian. *New Zealand Veterinary Journal* 2002; **50**: 36–38.
9. **Cousins DV, Dawson DJ.** Tuberculosis due to *Mycobacterium bovis* in the Australian population: cases recorded during 1970–1994. *International Journal of Tuberculosis and Lung Disease* 1999; **3**: 715–721.
10. **Hardie RM, Watson JM.** *Mycobacterium bovis* in England and Wales: past, present and future. *Epidemiology and Infection* 1992; **109**: 23–33.
11. **Maclean FS.** *Challenge for health: a history of public health in New Zealand.* Wellington: Government Printer, 1964.
12. **Thornley CN, et al.** Changing epidemiology of human leptospirosis in New Zealand. *Epidemiology and Infection* 2002; **128**: 29–36.
13. **van Soolingen D, et al.** Use of various genetic markers in differentiation of *Mycobacterium bovis* strains from animals and humans and for studying epidemiology of bovine tuberculosis. *Journal of Clinical Microbiology* 1994; **32**: 2425–2433.
14. **Szewzyk R, et al.** Molecular epidemiological studies of *Mycobacterium bovis* infections in humans and animals in Sweden. *Journal of Clinical Microbiology* 1995; **33**: 3183–3185.
15. **LoBue PA, et al.** Identification of a familial cluster of pulmonary *Mycobacterium bovis* disease. *International Journal of Tuberculosis and Lung Disease* 2004; **8**: 1142–1146.
16. **Fritsche A, et al.** *Mycobacterium bovis* tuberculosis: from animal to man and back. *International Journal of Tuberculosis and Lung Disease* 2004; **8**: 903–904.
17. **LoBue PA, LeClair JJ, Moser KS.** Contact investigation for cases of pulmonary *Mycobacterium bovis*. *International Journal of Tuberculosis and Lung Disease* 2004; **8**: 868–872.