Changing prevalence of tuberculosis infection with increasing age in high-burden townships in South Africa


*Desmond Tutu HIV Centre, Institute of Infectious Disease and Molecular Medicine, University of Cape Town, Cape Town, South Africa; † Developmental Center for AIDS Research and ‡ Department of Biostatistics and Computational Biology, University of Rochester School of Medicine and Dentistry, New York, New York, USA; § Institute of Infectious Diseases and Molecular Medicine, University of Cape Town, Cape Town, South Africa; § Division of Medicine, Imperial College, London, ** National Institute for Medical Research, London, * National Institute of Infectious and Tropical Diseases, London School of Hygiene & Tropical Medicine, London, UK

**Correspondence to:** Stephen D Lawn, Desmond Tutu HIV Centre, Institute of Infectious Disease and Molecular Medicine, Faculty of Health Sciences, University of Cape Town, Anzio Road, Observatory 7925, Cape Town, South Africa. Tel: (+27) 021 650 6957. Fax: (+27) 021 650 6963. e-mail: stevelawn@yahoo.co.uk

**Article submitted 7 September 2009. Final version accepted 20 October 2009.**

**SUMMARY**

**SETTING:** Crowded townships of Cape Town, South Africa, where human immunodeficiency virus (HIV) prevalence and tuberculosis (TB) notification rates are among the highest in the world.

**OBJECTIVES:** To determine age-specific prevalence rates of latent tuberculosis infection (LTBI) among HIV-negative individuals, and the annual risk and force of infection during childhood and adolescence.

**DESIGN:** A cross-sectional survey using a standardised tuberculin skin test (TST) in HIV-negative individuals aged 5–40 years. A TST diameter of $\geq 10$ mm was defined as indicative of LTBI.

**RESULTS:** Among 1061 individuals, only 4.7% had low-grade TST responses of 1–9 mm. However, the proportions of individuals with TST $\geq 10$ mm increased from 28.0% in the 5–10 year age stratum to 88.0% in the 31–35 year age stratum. The mean annual risk of infection was 3.9% up to 5 years of age. The estimated force of infection (the rate of acquisition of LTBI among the residual pool of non-infected individuals) increased throughout childhood to a maximum of 7.9% per year at age 15 years.

**CONCLUSIONS:** Extremely high rates of infection in childhood and adolescence result in very high LTBI prevalence rates in young adults who are most at risk of acquiring HIV infection. This may be an important factor fuelling the high rates of HIV-associated TB in southern Africa.

**KEY WORDS:** TB infection; age; sex; Africa; TST

**SOUTH AFRICAN tuberculosis (TB) notifications have increased six-fold over the last two decades, largely as a result of increasing human immunodeficiency virus (HIV) prevalence.**¹ A total of 461 000 new cases of TB in 2007 reflected one of the highest national TB notification rates in the world. The overall incidence rate was estimated at 948 per 100 000 population, of which 73% were estimated to be co-infected with HIV. South Africa alone accounted for approximately 25% of the global burden of HIV-associated TB.¹

Total TB notifications in Cape Town, a city of 3.2 million people, reached 27 000 in 2006.² The distribution of TB cases within this population, however, is very unequal, with unprecedentedly high burdens in the crowded and socially deprived African townships. Here TB annual notification rates were reported to exceed 1500/100 000 in 2006.³⁻⁵ Fuelled by high HIV prevalence, TB notifications are now most frequent between the ages of 20 and 40 years.⁶

This marked deterioration of TB control in South Africa over the past two decades of increasing HIV prevalence has occurred despite reported progress towards National TB Control Programme case management targets.¹⁷ Coverage using the World Health Organization (WHO) DOTS strategy is 100% and case detection rates have remained above target since 2003.¹ In the decade from 1996 to 2005, treatment default and treatment failure during treatment improved respectively from 18.1% to 10.4% and from 3.5% to 1.7%.⁸

In 1990, Karel Styblo, a leading protagonist of effective case management for TB control, postulated that ‘the impact of HIV infection on the epidemiological situation of TB is so large that under some conditions, the tools available at present for TB control
will fail to restrain the incidence of TB caused by HIV infection.\(^9\) He further proposed that the impact of HIV infection would depend not only on the prevalence of HIV infection but also on the prevalence of latent TB infection (LTBI) and the annual risk of TB infection (ARTI) in the general population. However, although the HIV epidemic in South Africa has been carefully monitored by annual seroprevalence surveys among antenatal women and by household surveys,\(^7,10\) few data describe the prevalence of LTBI or the ARTI in the general population.

We therefore undertook a study to measure the prevalence of LTBI in the township populations of Cape Town. The tuberculin skin test (TST) has been the most widely used of all the immunological tests for estimation of prevalence, incidence and trend of \textit{Mycobacterium tuberculosis} infection in populations, despite concerns about its sensitivity and specificity.\(^11\) We assessed TST responses among healthy HIV-negative residents of high-density townships aged between 5 and 40 years and used these to derive estimates of the age-specific prevalence of LTBI. Further analyses estimated the ARTI in the study population and the force of TB infection (the rate of acquisition of TB infection among the residual pool of non-infected individuals). These data provide important insights into the explosive impact of the high rates of HIV acquisition in young adults on TB control.

**METHODS**

**Study population**

TST responses were assessed in 1061 healthy individuals, including children and adolescents aged 5–17 years (\(n = 832\)) and HIV-negative adults aged 18–40 years (\(n = 229\)). Participants were residents of high-density predominantly black townships of Cape Town, where annual TB notification rates in 2006 exceeded 1500/100 000.\(^3–5\) Children were all school attendees, as described in a previous study,\(^3\) and healthy HIV-negative adults were recruited from a prospective cohort recruited for a Phase III HIV vaccine study (\(n = 60\)) or from HIV voluntary counselling and testing centres (\(n = 169\)). Eligibility criteria included residence within the study communities described above, age between 5 and 40 years, confirmed HIV-negative (adults only), non-pregnant, no previous or currently suspected episodes of TB and no exposure to isoniazid preventive therapy or corticosteroids.

These studies were approved by the University of Cape Town Research Ethics Committee.

**Tuberculin skin testing**

TST responses were assessed using the WHO-recommended standard methodology.\(^12\) Two tuberculin units of purified protein derivative (PPD) RT23 (Statens Serum Institut, Copenhagen, Denmark) were administered intradermally to the volar surface of the left forearm. Induration was assessed between 48 h and 72 h after inoculation and the diameter was measured. Age and sex were also recorded for each participant, as was bacille Calmette-Guérin (BCG) scar status in children.

**Definition of latent TB infection**

TST induration of \(\geq 10\) mm at the inoculation site was considered indicative of LTBI.

**Statistical analysis**

‘R’ statistical software was used for analyses. The relationship between TST diameter and age was explored by regression analysis. A generalised non-parametric logistic model\(^13\) was proposed to quantify the relationship between LTBI prevalence rate (as defined by TST diameter \(\geq 10\) mm) and age. We fitted the model using the penalised regression spline approach.\(^14\) To evaluate sex differences, prevalence rates for female and male subjects were obtained separately using a generalised semi-parametric logistic model which was then fitted to the data.\(^15\) The proportional rate of change in TB prevalence per year increase in age was estimated using the derivative of the prevalence rate with respect to the age of subjects. The annual risk of infection was calculated as: \(1 - (1 - \text{prevalence})^{1/\text{mean age} + 0.5}\). The force of infection was also calculated at specific ages for the pool of individuals who remained non-infected (annual change in prevalence/[1 – prevalence]).

**RESULTS**

**TST diameters**

TST responses were available from 1061 individuals, 524 male and 537 female. Diameters of induration ranged between 0 and 35 mm, with a mode within the 16–20 mm stratum (Figure 1). No TST response was recorded in 535 individuals (50.4%), 1–5 mm in
8 individuals (0.8%), 5–9 mm in 41 individuals (3.9%) and ⩾10 mm in 477 (45%) individuals (Figure 1).

To establish whether the magnitude of TST responses was associated with age, TST diameters recorded as ⩾1 mm were plotted against age (Figure 2). The linear regression line for these data approximated to a horizontal line, indicating no overall significant relationship. Furthermore, as previously reported, there was no significant difference in mean TST diameter among those aged 5–17 years when comparing those who had observable BCG scars (n = 213) and those who did not (P = 0.28).

Prevalence of LTBI

Using a TST diameter of ⩾10 mm to define LTBI, the age-stratified prevalence of infection for the total population and for male and female sub-populations was calculated (Table 1). LTBI prevalence increased steadily from 28.0% in the 5–10 years age stratum to a peak of 88.2% in the 31–35 years age stratum.

To better demonstrate the quantitative relationship between LTBI prevalence and age, a non-parametric curve fitting approach was used (Figure 3). The smoothed curve shows best fit of the prevalence with increasing age together with 95% confidence intervals (95%CIs). This analysis was also used to derive estimates of the LTBI prevalence at specific ages (Table 2). This shows that LTBI was estimated to be present in approximately one third of 10-year-old children, one half of adolescents aged 15 years, two thirds of 20-year-olds and three quarters of 25-year-olds (Table 2).

LTBI prevalence among males and females

Estimates of LTBI prevalence were separately derived for male and female subjects (Figure 4). The difference in prevalence rates between the male and female subjects was greatest among the older study participants. The maximum estimated LTBI prevalence among males was at 33 years (85%) compared to...
29 years among females (78%). The decreasing trend in prevalence at ages >29 years was mainly due to changes in prevalence among females. However, the main sex effect ($P = 0.22$) and the age-sex interaction effect ($P = 0.29$) were not statistically significant.

**Rates of change in LTBI prevalence by age**

Estimates of the change in LTBI prevalence rate per year increment in age were derived for males and females (Figure 5). The patterns of the curves were similar for both groups. The rate of change in prevalence rate increased from 5 years of age and peaked at the age of 13 years. Maximum rates were 4.4% per year in males and 3.7% per year in females (Figure 5). After age 13 years, the annual rate of change in LTBI prevalence decreased.

**Annual risk and force of infection**

Further analyses estimated the ARTI among children aged 5, 10 and 15 years, showing a range of 3.9% to 4.8% per year (Table 2). The force of infection was also calculated at specific ages for the pool of individuals who remained non-infected (annual change in prevalence/[1 – prevalence]; Table 2). This parameter reached a peak of 7.9% (95%CI 2.5–13.2) per year among individuals aged 15 years and was negative above the age of 30 years. This analysis of force of infection did not adjust for active TB disease (an exclusion criterion from the cohort) or TB-associated mortality.

**DISCUSSION**

We have shown rapidly increasing prevalence of TST responses in healthy HIV-negative township residents aged between 5 and 40 years. Using a cut-off of $\geq$10 mm diameter of induration as evidence of LTBI, we found that, by the age of school entry, almost a fifth of children were already infected. By the average age of sexual debut at 15 years,16 50% of adolescents in these communities were infected. By the age of 25 years, when HIV prevalence peaks in South Africa,7 approximately 75% of individuals had evidence of LTBI. The rate of increase in the prevalence of LTBI was maximal at 13 years of age in both males and females. Between the ages of 5 and 15 years, the mean ARTI remained exceptionally high (range 3.9–4.8%), while the force of infection (the risk of infection in the residual pool of non-infected individuals) was maximal, at 7.8%, at the age of 15 years.

The complexities and limitations of the TST have been extensively discussed elsewhere.11,12,17 Controversies have included the choice of the antigen utilised, the threshold for defining positivity and the performance characteristics of the test in different settings. The tuberculin reagent employed in this study was standard WHO-recommended PPD RT23, which allows comparison with other population surveys.18 Non-specific sensitivity resulting from exposure to environmental mycobacteria impairs test performance in some populations,11 manifesting with a high proportion of low-positive results. In our study population, there was a relatively clear separation between negative and positive results; the mode of positive results was in the 16–20 mm diameter stratum and the proportion of low positive results was very small. Furthermore, diameter of induration was not directly associated with known confounders, such as presence of BCG scar or the age of participants. We therefore used the conventional cut-off for test positivity of $\geq$10 mm.
The relationship between prevalence of LTBI, age and sex is determined by the individual's current and lifetime exposure to infection. While prevalence in children reflects more recent transmission, prevalence in adults reflects overall historical trends in transmission risk. It has been observed that, at low prevalence rates of LTBI, instability of TST reactions may lead to false interpretations of secular trends. However, the predictive value of TST responses is much greater when prevalence of infection is high.11 In our study population, TST performance characteristics appeared good and LTBI prevalence rates were exceptionally high. Furthermore, trends in sputum smear-positive TB notifications are very well documented, increasing relentlessly over the last two decades in South Africa and particularly in the study communities.1,6,20

To explore the relationship between LTBI prevalence, age and sex, we fitted a generalised non-parametric (i.e., no assumptions about the data distribution) logistic model. The resultant ‘S’-shaped curve had strong similarities with age and sex prevalence curves from TST surveys performed over 50 years ago in urban and rural sites of Bechuanaland (Botswana), Ghana, Liberia, Nigeria, Sierra Leone and Swaziland.21

Between 5 and 15 years of age, the curve is concave upwards (Figure 3). This could be indicative of decreasing transmission in recent years22 but, in the known context of rapidly increasing TB notifications over the past two decades, this is much more likely to be indicative of increasing risk of infection with age.23 The finding of maximal risk of acquisition in the mid-teenage years may reflect social mixing patterns and associated TB exposure in this age group.23,24

From 15 to 30 years, the curve had an exponential form (Figure 3), compatible with either a steady or decreasing infection risk with age.23 From 30 years onwards, declining prevalence with age was more marked in females. Declining of immunological memory with age did not appear to be a significant contributor, as reaction size was not shown to diminish significantly with age. Alternative explanations could include immigration from lower prevalence settings, survival from a period of lower TB transmission or preferential progression of individuals with TST positivity to TB disease, as a history of TB treatment was an exclusion criterion in adult participants.

Larger TST studies, such as those performed between 1955 and 1960, included heterogeneous populations with very variable TB exposure rates.23 This study, which was modest in size (n = 1061), focused particularly on a population within a single city that has an extraordinary high burden of HIV and TB disease. The study population was not representative of the total community, as HIV-infected individuals and those who had already developed TB disease were excluded. We believe that the risk of TB infection and performance characteristics of TST in HIV-infected patients and patients with TB disease may be very different. The results are therefore only descriptive of those remaining free of both TB and HIV. The number of individuals aged >30 years was relatively small, leading to wide CIs around prevalence estimates in this age group. The differences between calculated and modelled prevalence estimates (Table 1 and Figure 3) were not statistically significant.

In summary, we have shown an extremely high rate of acquisition of LTBI in childhood and adolescence in poor African townships of Cape Town. The rates observed are as high as the highest rates observed in the WHO surveys conducted in African countries over 50 years ago, in the era prior to modern multidrug TB treatment and TB control programmes.21 The combination of high prevalence and force of infection in adolescence before the acquisition of HIV infection may be a key factor underlying the explosive HIV-associated TB epidemic in South Africa. HIV prevalence among those aged 20–39 years in these communities reached 30% in 2002,2 and the current data suggest that approximately two thirds of these individuals were likely to have already been infected with M. tuberculosis infection prior to HIV acquisition. This may be an important factor fuelling high rates of HIV-associated TB in southern Africa.

The long-term aim of TB control is to lower infection rates in successive generations. Present facility-based TB control is failing to reduce TB infection rates in children and adolescents in these communities. Systematic evaluation and reduction of infection rates in these high-burden communities of Southern Africa should be incorporated as a target of TB control.

Acknowledgements
This research was a collaborative project of the Developmental Centre for AIDS Research (D-CFAR), University of Rochester, NY, USA. RW and LGB are funded in part by the National Institutes of Health (NIH/NAID) grants 1U19AI53217-01 (RW and LGB) and by grant R01 A1058736-01A1 (RW). HL is funded by NIH/NAID grants AI59773 and National Science Foundation grant DMS 0806097. HW is funded by NIH/NAID grants AI50020 and
CONTEXTE : Les faubourgs peuplés de Cape Town, Afrique du Sud, où les taux de déclaration de la prévalence du virus de l’immunodéficience humaine (VIH) et de la tuberculose (TB) sont parmi les plus élevés du monde.

OBJECTIF : Déterminer les taux de prévalence de l’infection tuberculose latente (LTBI) spécifiques pour l’âge parmi les individus séronégatifs pour le VIH.

SCHEMA : Enquête transversale utilisant un test tuberculine standardisé (TST) chez les individus séronégatifs pour le VIH âgés de 5 à 40 ans. On a défini comme indiquant la LTBI un diamètre du TST ≥ 10 mm.

RESULTATS : Parmi 1061 individus, 4,7% seulement ont eu des réactions du TST de faible degré (1 à 9 mm). Toutefois, les proportions d’individus dont les diamètres de TST sont ≥ 10 mm augmentent de 28% dans la classe d’âge de 5 à 10 ans jusqu’à 88% dans la classe d’âge de 31 à 35 ans. Le risque annuel moyen d’infection est de 3,9% jusqu’à l’âge de 5 ans. La puissance estimée de l’infection (taux d’acquisition de la LTBI dans le réservoir résiduel d’individus non-infectés) augmente pendant l’enfance jusqu’à un maximum de 7,9% par an à l’âge de 15 ans.

CONCLUSIONS : Les taux extrêmement élevés d’infection dans l’enfance et l’adolescence entraînent les taux très élevés de prévalence de la LTBI chez les jeunes adultes où le risque de l’infection VIAH est maximal. Ceci peut être un facteur important qui nourrit les taux élevés de tuberculose associée au VIH en Afrique du Sud.
MARCO DE REFERENCIA: La prevalencia de infección por el virus de la inmunodeficiencia humana (VIH) y las tasas de notificación de tuberculosis (TB) en las localidades densamente pobladas de la Ciudad del Cabo en Sudáfrica, se encuentran entre las más altas del mundo.

OBJETIVOS: Determinar las tasas de prevalencia específicas por edad de infección tuberculosa latente (LTBI) en las personas con examen serológico negativo para el VIH y evaluar el riesgo anual y la fuerza de la infección durante la niñez y la adolescencia.

MÉTODOS: Se llevó a cabo un estudio transversal aplicando la prueba cutánea de la tuberculina (TST) a personas entre los 5 y los 40 años de edad, con serología negativa para el VIH. Se escogió una reacción de diámetro \( \geq 10 \text{ mm} \) como indicativo de LTBI.

RESULTADOS: De las 1061 personas que participaron, solo 4,7% tuvieron reacciones menores a la TST (con diámetros de 1 mm a 9 mm). Sin embargo, la proporción de personas con reacciones de diámetro \( \geq 10 \text{ mm} \) aumentó de 28,0% en el grupo de 5 a 10 años de edad hasta 88,0% en el grupo de 31 a 35 años de edad. El riesgo anual de infección promedio fue 3,9% hasta los 5 años de edad. La fuerza calculada de la infección (la tasa de contracción de la LTBI en un grupo residual de personas sin infección) aumentó durante la infancia, hasta un máximo de 7,9% por año a la edad de 15 años.

CONCLUSIONES: Las tasas extremadamente altas de infección tuberculosa en la infancia y la adolescencia conllevan altas tasas de prevalencia de LTBI en los adultos jóvenes, quienes presentan el mayor riesgo de contraer la infección por el VIH. Este puede ser un factor importante que alimenta las altas tasas de tuberculosis asociada con la infección por el VIH en Sudáfrica.