Extensively drug-resistant tuberculosis as a cause of death in patients co-infected with tuberculosis and HIV in a rural area of South Africa

Neel R Gandhi, Anthony Moll, A Willem Sturm, Robert Pawinski, Thiloshini Govender, Umesh Lalloo, Kimberly Zeller, Jason Andrews, Gerald Friedland

Summary

Background **The epidemics of HIV-1 and tuberculosis in South Africa are closely related. High mortality rates in co-infected patients have improved with antiretroviral therapy, but drug-resistant tuberculosis has emerged as a major cause of death. We assessed the prevalence and consequences of multidrug-resistant (MDR) and extensively drug-resistant (XDR) tuberculosis in a rural area in KwaZulu Natal, South Africa.**

Methods **We undertook enhanced surveillance for drug-resistant tuberculosis with sputum culture and drug susceptibility testing in patients with known or suspected tuberculosis. Genotyping was done for isolates resistant to first-line and second-line drugs.**

Results **From January, 2005, to March, 2006, sputum was obtained from 1539 patients. We detected MDR tuberculosis in 221 patients, of whom 53 had XDR tuberculosis. Prevalence among 475 patients with culture-confirmed tuberculosis was 39% (185 patients) for MDR and 6% (30) for XDR tuberculosis. Only 55% (26 of 47) of patients with XDR tuberculosis had never been previously treated for tuberculosis; 67% (28 of 42) had a recent hospital admission. All 44 patients with XDR tuberculosis who were tested for HIV were co-infected. 52 of 53 patients with XDR tuberculosis died, with median survival of 16 days from time of diagnosis (IQR 6–37) among the 42 patients with confirmed dates of death. Genotyping of isolates showed that 39 of 46 (85%, 95% CI 74–95) patients with XDR tuberculosis had similar strains.**

Conclusions **MDR tuberculosis is more prevalent than previously realised in this setting. XDR tuberculosis has been transmitted to HIV co-infected patients and is associated with high mortality. These observations warrant urgent intervention and threaten the success of treatment programmes for tuberculosis and HIV.**

Lancet 2006; 368: 1575–80

Published Online October 26, 2006 DOI:10.1016/S0140-6736(06)69573-1

See Comment page 1554

AIDS Program, Section of Infectious Diseases, Department of Internal Medicine, Yale University School of Medicine, New Haven, CT, USA (N R Gandhi MD, J Andrews BA, K Zeller MD, G Friedland MD); **Church of Scotland Hospital and Philanjalo, Tugela Ferry, South Africa** (A Moll MBChB); **Department of Medical Microbiology** (A W Sturm MD) **and Department of Medicine** (R Pawinski MBChB, U Lalloo MD), **Nelson R Mandela School of Medicine, Durban, South Africa; KwaZulu Natal Department of Health, KwaZulu Natal, South Africa** (T Govender MBChB); **Brown University School of Medicine, Providence, RI, USA** (K Zeller), **and Department of Medicine, Division of General Internal Medicine, Albert Einstein College of Medicine, Bronx, NY 10467, USA** (N R Gandhi)

Correspondence to: Dr Neel R Gandhi **negandhi@montefiore.org**

Introduction

Tuberculosis is the most common cause of morbidity and mortality in individuals with HIV-1 infection in sub-Saharan Africa.[1] HIV greatly increases the risk of active tuberculosis disease[2] and about 80% of patients presenting with active tuberculosis in the province of KwaZulu Natal, South Africa, are co-infected with HIV. Mortality rates of up to 40% per year have been reported in patients co-infected with tuberculosis and HIV who are receiving treatment for tuberculosis, but not for HIV.[3] Although antiretroviral therapy is likely to reduce HIV-associated morbidity and mortality as it becomes more widely available, any reduction is likely to be blunted if efforts are not taken to improve tuberculosis programmes concurrently.[4–6]

The number of tuberculosis cases in sub-Saharan Africa has increased substantially in the past decade, fuelled by the HIV epidemic,[7] making it difficult for tuberculosis programmes to improve outcomes.[8] In South Africa, the national DOTS treatment success rate has been reported to be only 67%,[9] well below the WHO standard of 85%.[10] Low rates of treatment completion place patients at risk for relapse of tuberculosis disease as well as for development of drug-resistance.

Rates of multidrug-resistant (MDR) tuberculosis among new cases of tuberculosis in sub-Saharan Africa have been low in the past, ranging from 0·8% to 2·6% in the last global drug resistance survey (1999–2002), compared with 7·8–14·2% in countries with the highest rates.[11] However, the prevalence of drug resistance in the region seems to have risen since the last global drug resistance survey.[12,13] In KwaZulu Natal, South Africa, the rate of MDR tuberculosis in new patients was reported at 1·7% between 2000 and 2002;[14] the rate was 9% in a study integrating treatment for tuberculosis and HIV that we undertook from 2003 to 2006 in the same region.[15]

Resistance to second-line treatment for tuberculosis is another concern that has recently been raised in a study by Shah and colleagues.[16] The investigators found emerging resistance not only to isoniazid and rifampicin (MDR tuberculosis), but also to at least three classes of second-line drugs, which they termed extensively drug-resistant (XDR) tuberculosis. 347 patients with the disease were described worldwide,[16] but data from Africa were few and data about HIV co-infection were not available. In our tuberculosis-HIV integration study in KwaZulu Natal, six of 119 (5%) patients co-infected with both diseases met the revised criteria for XDR

tuberculosis,[17] defined as resistance to at least isoniazid, rifampicin, fluoroquinolones, and either aminoglycosides (amikacin, kanamycin) or capreomycin, or both. This finding raised the concern that not only MDR tuberculosis, but also XDR tuberculosis, was emerging in this region with high HIV prevalence.[15]

We therefore undertook a study to assess the extent of MDR tuberculosis and XDR tuberculosis in this rural area in South Africa. We also aimed to describe characteristics and treatment histories of individuals with XDR tuberculosis in this setting.

Methods

Setting and study population

We did this study in the Msinga sub-district of KwaZulu Natal, South Africa, a 2000 km^2 rural area, which is home to 300 000 traditional Zulu people. A provincial government district hospital of 355 beds is the focus of health care for this population. 40% of inpatient beds in this hospital are occupied by patients infected with HIV, and the prevalence of HIV infection in women presenting to the maternity ward is 20%. A government-sponsored tuberculosis treatment programme, using the WHO DOTS strategy,[18] has been in place in this district since 1993. Patients receive free treatment for tuberculosis by home-based directly observed therapy, administered by volunteer community health workers. The standard regimen, regardless of HIV status, is isoniazid, rifampicin, ethambutol, and pyrazinamide for 2 months, followed by 4 months of isoniazid and rifampicin. Diagnosis is typically made by sputum microscopy for acid-fast bacilli, x-ray, or clinical criteria, according to the South African National Tuberculosis Guidelines.[19] Sputum cultures are not routinely done in patients suspected of having tuberculosis for the first time, but are done in those with treatment failure or recurrence. Patients identified as having MDR tuberculosis are referred to a dedicated tuberculosis hospital in metropolitan Durban, where most second-line drugs are available (at the time of the study, capreomycin and para-aminosalicylic acid were not available in South Africa).

A government-sponsored antiretroviral therapy programme was started at this study site in March, 2004. Patients with CD4-positive T-lymphocyte (CD4) counts of less than 200 cells per mm^3 were eligible for free antiretroviral treatment. By September, 2006, nearly 1300 patients had been enrolled from this site.

Patients were included in this study from January, 2005, to March, 2006, and divided into three groups. Group 1 included consecutive patients in whom a tuberculosis culture was done in accord with the South African guidelines[19] between January and May, 2005. This group consisted of individuals with persistently smear-positive sputum specimens and those with recurrent tuberculosis. Group 2 included all inpatients present on the male and female tuberculosis wards on a single day in February, 2005. Group 3 consisted of consecutive inpatients and outpatients who presented with signs and symptoms of tuberculosis (eg, cough, fever, weight loss) at the district hospital between June, 2005, and March, 2006.

Procedures

Sputum samples were obtained from all patients for mycobacterial culture and drug susceptibility testing. Typically, one to three samples were taken per patient. The samples were not induced and were taken at any time of day. Sputum specimens were stored at 4°C for up to 3 days until transport to the provincial diagnostic mycobacteriology laboratory in Durban. Digestion and decontamination was done with the N-acetyl-L-cysteine-sodium hydroxide method. An auramine-stained smear was made and the remaining deposit was inoculated in one mycobacteria growth indicator tube (MGIT) broth and on one Middlebrook 7H10 agar plate. The broths were incubated at 37°C in an automated incubator. Agar plates were sealed in CO_2-permeable plastic bags and incubated in 5% CO_2 at 37°C. Acid-fast microscopy was done on each positive MGIT broth when a positive reading was obtained. Those containing acid-fast bacilli were subcultured on Middlebrook 7H10 agar. Primary Middlebrook agar plates were read weekly for 3 weeks or until growth was observed. Microscopy was done to confirm the presence of acid-fast bacilli. All positive cultures were identified as *Mycobacterium tuberculosis* by means of niacin and nitrate reductase tests.

The risk of cross-contamination was minimised by processing samples individually in real time, rather than batching. Quality assurance was done weekly by the UK National External Quality Assessment Service programme, where ten consecutive isolates were fingerprinted to rule out cross-contamination.

Susceptibility tests were done on all isolates using the 1% proportional method on Middlebrook 7H10 agar. All isolates were tested for susceptibility to isoniazid (1 mg/L), rifampicin (2 mg/L), ethambutol (5 mg/L), streptomycin (2 mg/L), kanamycin (16 mg/L) and ciprofloxacin (2 mg/L). Susceptibility testing to pyrazinamide and the remaining four classes of second line drugs—ethionamide, cycloserine, capreomycin, and para-aminosalicylic acid—are not routinely done.

Genotyping by IS6110 fingerprinting[20] and spoligotyping[21] was done on isolates found to have resistance to first-line and second-line drugs. IS6110 fingerprints were analysed by GelCompar 4.0 software (Applied Maths, Kortrijk, Belgium); spoligotyping patterns were analysed by visual inspection. Strains were classified as belonging to the KwaZulu Natal (KZN) family of tuberculosis strains if there was a difference of two bands or less by IS6110 fingerprinting, or if fewer than five spacers were absent compared with the typical KZN pattern by spoligotyping.

Definitions, analysis, and outcomes

Positive cultures for *M tuberculosis* were categorised on the basis of drug susceptibility results, as: fully susceptible or resistant to one or more tuberculosis drugs, but not

both isoniazid and rifampicin (non-MDR tuberculosis); resistant to at least both isoniazid and rifampicin (MDR tuberculosis); or resistant to at least isoniazid, rifampicin, fluoroquinolones, and either aminoglycosides (amikacin, kanamycin) or capreomycin, or both (XDR tuberculosis). We calculated prevalence rates of MDR and XDR tuberculosis among confirmed tuberculosis cases (patients with positive tuberculosis cultures) in group 3. The probability of having MDR or XDR tuberculosis in patients presenting to this district hospital with signs or symptoms of tuberculosis was also calculated in group 3.

We reviewed hospital medical records for all cases of XDR tuberculosis to determine patients' demographics, previous tuberculosis treatment, previous hospital admission, HIV history, and vital status. Tuberculosis treatment history was classified by standard definitions: cure, treatment completion, treatment failure, default, transferred out, or death.[22] For patients tested for HIV, information about most recent CD4 count and viral load was obtained, as well as any information about antiretroviral therapy.

The primary outcomes of interest were number of cases and prevalence rates of MDR and XDR tuberculosis. Secondary outcomes for patients with XDR tuberculosis were mortality, proportion with previous treatment for tuberculosis, proportion with previous hospital admission, HIV co-infection, and genotype of isolates. Differences in the duration of survival on the basis of patient's characteristics and previous treatment were analysed with ANOVA.

The study was approved by the Ethics and Human Investigation Committees of the University of KwaZulu Natal and Yale University.

Role of the funding source

The sponsors of the study had no role in study design, data collection, data analysis, data interpretation, or writing of the report. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit for publication.

Results

We sent 2203 sputum specimens from 1539 individual patients for mycobacterial culture between Jan 1, 2005, and March 31, 2006. 542 patients had at least one culture that was positive for *M tuberculosis*. Table 1 shows the distribution of these patients and the classification of resistance over the three groups. In total, 221 cases of MDR tuberculosis were identified. Of these, 53 patients had XDR tuberculosis, with resistance to isoniazid, rifampicin, ethambutol, streptomycin, aminoglycosides, and fluoroquinolones.

Of the 1428 patients presenting to this district hospital with signs and symptoms of tuberculosis (group 3), 475 (33%) had active culture-positive tuberculosis (table 1). Among these confirmed cases, the prevalence of MDR tuberculosis was 39% (185) and of XDR tuberculosis was 6% (30). Among all patients presenting with signs and symptoms of tuberculosis, the probability of having MDR tuberculosis was 13% (185 of 1428) and XDR tuberculosis was 2% (30 of 1428).

Of the 53 patients with XDR tuberculosis, 25 (49%) were women and the median age was 35 years (range 20–75 years; table 2). Data on tuberculosis characteristics, previous treatment for tuberculosis, history of hospital admission, and HIV characteristics are presented in table 2. Notably, the majority of patients (55%) had never previously received treatment for tuberculosis, while an additional 30% had documented cure or completion of their previous tuberculosis treatment course. 67% of patients had been admitted to this district hospital for any cause in the 2 years preceding their presentation with XDR tuberculosis. Two patients with XDR tuberculosis were health-care workers in the hospital; both died of XDR tuberculosis. Four other hospital workers had been suspected of having the condition, but sought care at another hospital and as a result were not included in this cohort.

Contact tracing was completed for all 53 patients with XDR tuberculosis. They were from a dispersed geographical region with no known contact with each other apart from receiving health care from the same district hospital. None of the patients had a family member who was sick with tuberculosis before his or her illness.

	Group 1	Group 2	Group 3†	Total
Total tested	86	25	1428	1539
Culture-positive	45	22	475	542
MDR tuberculosis*	26	10	185	221
XDR tuberculosis	17	6	30	53

Data are number of patients. *Includes cases of XDR tuberculosis.

Table 1: **Distribution of culture results and drug-resistance categories by group for all patients (n=1539) for whom sputum culture was done**

	Number (%)
Tuberculosis characteristics (n=53)	
Pulmonary tuberculosis alone	40 (75%)
Pulmonary and extrapulmonary tuberculosis	13 (25%)
Sputum-smear positive	42 (79%)
Sputum-smear negative	11 (21%)
Previous tuberculosis treatment (n=47)	
No previous treatment	26 (55%)
Previous treatment: cure or completed treatment	14 (30%)
Treatment default or failure	7 (15%)
Previous admission in past 2 years (n=42)	
Admitted for any cause	28 (67%)
No previous admission	14 (33%)
HIV characteristics (n=44)	
HIV-infected	44 (100%)
On antiretroviral therapy	15 (34%)

Table 2: **Characteristics of patients with XDR tuberculosis**

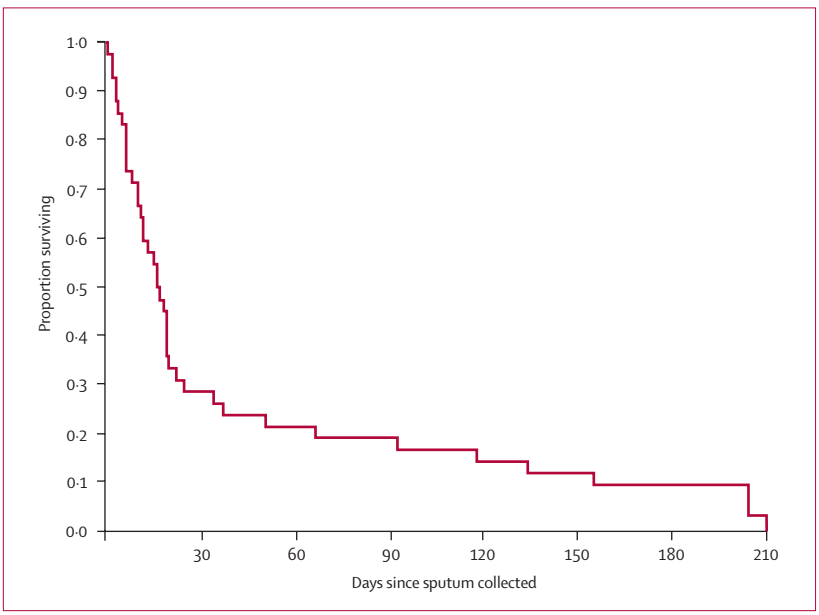

Figure: **Survival after sputum collection in patients with XDR tuberculosis with confirmed dates of death (n=42)**

All 44 patients with XDR tuberculosis who had been tested for HIV were infected with the virus (table 2). Their median CD4 count at the time of sputum collection was 63 cells per mm^3 (range 9–283). Viral loads on antiretroviral treatment were not available for any of these patients.

52 of the 53 (98%) patients with XDR tuberculosis died. Median survival from the time of specimen collection to death was 16 days (range 2–210 days, IQR 6–37) in the 42 patients with confirmed dates of death (the remainder died at home or outside of the community and their precise dates of death were not documented in the medical records). The figure shows that roughly 70% of patients died within 30 days from the time when their sputum was collected for culture. The duration of survival did not vary significantly on the basis of age, sex, data collection group, previous treatment for tuberculosis, previous hospital admission, HIV status, CD4 count, or use of antiretroviral drugs.

Genotyping has been completed for isolates from 46 patients with XDR tuberculosis to date. IS6110 fingerprinting was done on 16 of 23 patients in groups 1 and 2, and spoligotyping on all 30 patients in group 3. Genotyping showed that 39 of the 46 (85%; 95% CI 74–95) isolates tested were genetically similar, belonging to the KZN family of strains; 13 of 16 (81%) isolates in groups 1 and 2 and 26 of 30 (87%) in group 3.

Discussion

We undertook enhanced surveillance for drug-resistant tuberculosis by instituting routine mycobacterial culture and drug susceptibility testing on patients with suspected or diagnosed tuberculosis in a rural resource-limited setting, with a high prevalence of HIV, in South Africa. We found a substantially higher prevalence of XDR tuberculosis and MDR tuberculosis than previously reported.[14] All patients with XDR tuberculosis who had previously been tested for HIV were co-infected with the virus. XDR tuberculosis disease in this population was rapidly, and almost uniformly, fatal.

The findings of our study cause concern for several reasons, beyond the lethal nature of the disease. More than half the patients with XDR tuberculosis had never been previously treated for tuberculosis; an additional third had either been cured or had completed treatment for previous tuberculosis illness. With only 15% of patients having treatment failure or default, most patients were unlikely to have developed resistant tuberculosis as a consequence of unsuccessful treatment. Instead, transmission of XDR strains between individuals has probably occurred; this assumption is supported by the genotyping results. About 85% of the XDR isolates were from the KZN family of tuberculosis strains, which was first described in 1996.[23] At that time, the KZN strains were either fully susceptible or had resistance to only first-line tuberculosis drugs.[24] Resistance to second-line drugs was not seen until the past 2–3 years,[24] further supporting the notion of recent transmission of XDR tuberculosis to our patients.

It is also probable that transmission of the XDR tuberculosis strain occurred nosocomially. We found that two-thirds of patients were recently hospitalised before the onset of XDR tuberculosis and that two health-care workers, and possibly four others, died from XDR tuberculosis. These findings are reminiscent of MDR tuberculosis outbreaks worldwide in the past 20 years,[25–34] in which drug-resistant strains transmitted nosocomially were responsible for extensive mortality in HIV patients. These findings are particularly worrying for resource-limited settings similar to the site of this study, where roughly 40% of patients admitted to hospital are HIV-infected and effective infection control facilities and practices are extremely limited.[4,35]

Tuberculosis is the most common opportunistic infection and cause of death among HIV-infected patients in resource-limited settings.[36,37] Patients with HIV infections are particularly vulnerable to primary disease following infection with tuberculosis,[33] and therefore are at high risk of illness and mortality when exposed to drug-resistant tuberculosis strains. Although antiretroviral therapy has reduced the incidence of active tuberculosis,[38] HIV-infected patients on therapy still have a more than five-fold increased risk of developing tuberculosis compared with individuals without HIV infection.[39] Although the combination of tuberculosis treatment and antiretroviral therapy can improve mortality in co-infected patients,[15,40,41] it is less likely to do so in patients with drug-resistant tuberculosis.[15] Compared with first-line treatment, second-line treatment for MDR tuberculosis requires a longer course, is more toxic, more costly, and is not readily available in resource-

limited settings. Treatment success in XDR tuberculosis is very difficult as few active drugs remain. Thus, drug-resistant tuberculosis and its nosocomial transmission threaten the achievements of DOTS and antiretroviral scale-up programmes, which are now widely being implemented in resource-limited settings worldwide.

A rapid and comprehensive approach is essential to tackle this ominous situation. First, the full extent of MDR and XDR tuberculosis in areas of high HIV prevalence needs to be ascertained. Resources are needed to establish laboratory capacity capable of undertaking mycobacterial culture and drug susceptibility testing to facilitate timely diagnosis and to assess prevalence of drug resistance, in resource-limited settings.[11]

Second, tuberculosis treatment programmes must be strengthened to improve treatment completion rates and provide treatment for drug-resistant disease. Treatment completion rates in many resource-limited settings are well below the WHO standard of 85%.[10] These low rates promote the development of drug-resistant strains, which could then result in their transmission. Treatment programmes providing second-line therapy for drug-resistant tuberculosis are needed to save lives and to reduce the further spread of drug-resistant strains.[42]

Infection control facilities and practices are generally inadequate in most resource-limited settings.[4] This shortcoming has undoubtedly contributed to magnitude of the problem in the present study. Improvements in infection control facilities and practices are crucial to break the cycle of transmission of drug-resistant tuberculosis, in addition to protecting health-care workers, an increasingly scarce resource in Africa and other resource-limited settings.

Last, simpler diagnostic tools for detecting active tuberculosis and drug resistance must be developed for dissemination in resource-limited settings.[43,44] The current diagnosis of drug-resistant tuberculosis relies on mycobacterial culture and drug susceptibility testing, which are time consuming, labour intensive, and costly, when available. Accelerated development of new drugs is also essential. With currently available drugs, patients with XDR tuberculosis are left with few, if any, treatment options.

We recognise some limitations to our study. First, the investigation was done at a single site in rural South Africa. Although the full extent of MDR and XDR tuberculosis in South Africa and beyond is not known, growing evidence suggests that cases are not confined to a local cluster. The presence of MDR and XDR tuberculosis has been documented in 28 other hospitals in KwaZulu Natal,[24] all provinces in South Africa[45] and worldwide.[16] Second, our data are limited by the unavailability of drug susceptibility testing for all classes of second-line drugs in South Africa. The isolates in our study were resistant to all first-line drugs for which tests were done and the two most important classes of second-line drugs: aminoglycosides and fluoroquinolones.[46] Although susceptibility results from the remaining classes were not available, the rapid and near-complete mortality in this population shows the potential grave consequences of transmission of drug resistant tuberculosis strains in high HIV prevalence setting.

Third, duration of survival was calculated from time of sputum collection rather than from the time of initial tuberculosis diagnosis or time of treatment initiation, because the sputum culture represents the first confirmation of XDR tuberculosis infection. Culture data from earlier points in their illness were not available for all patients in groups 1 and 2, since routine culture at the time of initiation of anti-tuberculosis therapy is not recommended by South Africa tuberculosis control guidelines. Therefore, we were unable to ascertain whether patients were ill from XDR tuberculosis from the time of their initial diagnosis or treatment initiation, or whether they were later reinfected or superinfected with an XDR tuberculosis strain. Lastly, our data provide characterisation of patients with XDR tuberculosis only. Similar data for patients with MDR and drug-susceptible tuberculosis are necessary to help identify predictive factors for resistant disease.

Despite these limitations, this study provides disturbing new evidence of the presence and serious consequences of drug-resistant tuberculosis in a resource-limited area, with a high prevalence of HIV, and highlights the need for urgent local and international intervention.

Contributors

All authors contributed to the conception and design of the study, analysis and interpretation of data, and to drafting or critical revision of the article. Data were acquired by A Moll, A W Sturm, R Pawinski, J Andrews, and G Friedland.

Conflict of interest statement

We declare that we have no conflict of interest.

Acknowledgments

We thank the staff and patients who made this study possible. This study received funding from the Irene Diamond Fund, the Doris Duke Charitable Foundation, and Yale University. N R Gandhi and K Zeller received support from Robert Wood Johnson Clinical Scholars Program while at Yale University and from The Emory School of Medicine and The Clinton Foundation, respectively.

References

1 Mukadi YD, Maher D, Harries A. Tuberculosis case fatality rates in high HIV prevalence populations in sub-Saharan Africa. *AIDS* 2001; **15:** 143–52.

2 Selwyn PA, Hartel D, Lewis VA, et al. A prospective study of the risk of tuberculosis among intravenous drug users with human immunodeficiency virus infection. *N Engl J Med* 1989; **320:** 545–50.

3 Wilkinson D, Davies GR. The increasing burden of tuberculosis in rural South Africa—impact of the HIV epidemic. *S Afr Med J* 1997; **87:** 447–50.

4 De Cock KM, Chaisson RE. Will DOTS do it? A reappraisal of tuberculosis control in countries with high rates of HIV infection. *Int J Tuberc Lung Dis* 1999; **3:** 457–65.

5 Corbett EL, Marston B, Churchyard GJ, De Cock KM. Tuberculosis in sub-Saharan Africa: opportunities, challenges, and change in the era of antiretroviral treatment. *Lancet* 2006; **367:** 926–37.

6 De Cock KM, Marston B. The sound of one hand clapping: tuberculosis and antiretroviral therapy in Africa. *Am J Respir Crit Care Med* 2005; **172:** 3–4.

7 Raviglione MC, Harries AD, Msiska R, Wilkinson D, Nunn P. Tuberculosis and HIV: current status in Africa. *Aids* 1997; **11** (suppl B)**:** S115–23.

8 Friedland G, Abdool Karim S, Abdool Karim Q, et al. Utility of tuberculosis directly observed therapy programs as sites for access to and provision of antiretroviral therapy in resource-limited countries. *Clin Infect Dis* 2004; **38:** S421–28.

9 WHO. Global tuberculosis control: surveillance, planning, financing. WHO report 2006 (WHO/HTM/TB/2006.362). Geneva: World Health Organization, 2006.

10 WHO. Framework for effective tuberculosis control. WHO/TB/94.179. Geneva: World Health Organization, 1994.

11 WHO/IUATLD Global Project on Anti-Tuberculosis Drug Resistance Surveillance. Anti-tuberculosis drug resistance in the world: third global report. Geneva: World Health Organization, 2004.

12 Nelson LJ, Talbot EA, Mwasekaga MJ, et al. Antituberculosis drug resistance and anonymous HIV surveillance in tuberculosis patients in Botswana, 2002. *Lancet* 2005; **366:** 488–90.

13 Nunes EA, De Capitani EM, Coelho E, et al. Patterns of anti-tuberculosis drug resistance among HIV-infected patients in Maputo, Mozambique, 2002–2003. *Int J Tuberc Lung Dis* 2005; **9:** 494–500.

14 Weyer K, Lancaster J, Brand J, van der Walt M, Levin J. Survey of tuberculosis drug resistance in KwaZulu Natal 2001–2002. Pretoria: Medical Research Council Unit for TB Epidemiology and Interventions Branch, 2003.

15 Gandhi N, Moll A, Pawinski R, Zeller K, Lalloo U, Friedland G. Favorable outcomes of integration of TB and HIV treatment in a rural South Africa: the Sizonqoba study. Toronto: XVI International AIDS Conference, 2006 (abstr MOPE0181).

16 Anon. Emergence of *Mycobacterium tuberculosis* with extensive resistance to second-line drugs—worldwide, 2000–2004. *MMWR Morb Mortal Wkly Rep* 2006; **55:** 301–05.

17 WHO. Laboratory XDR-TB definitions. Geneva: Meeting of the Global XDR TB Task Force, 2006.

18 WHO. Treatment of tuberculosis: guidelines for national programmes. 3rd edn. WHO/CDS/TB/2003.313. Geneva: World Health Organization, 2003.

19 South African Tuberculosis Control Programme. The South African Tuberculosis Control Programme practical guidelines. Pretoria: Republic of South Africa Department of Health, 2000.

20 van Embden JD, Cave MD, Crawford JT, et al. Strain identification of *Mycobacterium tuberculosis* by DNA fingerprinting: recommendations for a standardized methodology. *J Clin Microbiol* 1993; **31:** 406–09.

21 Kremer K, van Soolingen D, Frothingham R, et al. Comparison of methods based on different molecular epidemiological markers for typing of *Mycobacterium tuberculosis* complex strains: interlaboratory study of discriminatory power and reproducibility. *J Clin Microbiol* 1999; **37:** 2607–18.

22 WHO. Treatment of tuberculosis: guidelines for national programmes. 3rd edn (English). WHO/CDS/TB/2003.313. Geneva: World Health Organisation, 2003.

23 Davies GR, Pillay M, Sturm AW, Wilkinson D. Emergence of multidrug-resistant tuberculosis in a community-based directly observed treatment programme in rural South Africa. *Int J Tuberc Lung Dis* 1999; **3:** 799–804.

24 Sturm AW. The UKZN "Outbreak". Expert consultation on drug-resistant tuberculosis. Johannesburg: MRC Unit for TB Epidemiology and Interventions Branch, 2006.

25 Ritacco V, Di Lonardo M, Reniero A, et al. Nosocomial spread of human immunodeficiency virus-related multidrug-resistant tuberculosis in Buenos Aires. *J Infect Dis* 1997; **176:** 637–42.

26 Nosocomial transmission of multidrug-resistant tuberculosis among HIV-infected persons—Florida and New York, 1988–1991. *MMWR Morb Mortal Wkly Rep* 1991; **40:** 585–91.

27 Frieden TR, Sterling T, Pablos-Mendez A, Kilburn JO, Cauthen GM, Dooley SW. The emergence of drug-resistant tuberculosis in New York City. *N Engl J Med* 1993; **328:** 521–26.

28 Punnotok J, Shaffer N, Naiwatanakul T, et al. Human immunodeficiency virus-related tuberculosis and primary drug resistance in Bangkok, Thailand. *Int J Tuberc Lung Dis* 2000; **4:** 537–43.

29 Rullan JV, Herrera D, Cano R, et al. Nosocomial transmission of multidrug-resistant Mycobacterium tuberculosis in Spain. *Emerg Infect Dis* 1996; **2:** 125–29.

30 Sacks LV, Pendle S, Orlovic D, Blumberg L, Constantinou C. A comparison of outbreak- and nonoutbreak-related multidrug-resistant tuberculosis among human immunodeficiency virus-infected patients in a South African hospital. *Clin Infect Dis* 1999; **29:** 96–101.

31 Hannan MM, Peres H, Maltez F, et al. Investigation and control of a large outbreak of multi-drug resistant tuberculosis at a central Lisbon hospital. *J Hosp Infect* 2001; **47:** 91–97.

32 Campos PE, Suarez PG, Sanchez J, et al. Multidrug-resistant *Mycobacterium tuberculosis* in HIV-infected persons, Peru. *Emerg Infect Dis* 2003; **9:** 1571–78.

33 Daley CL, Small PM, Schecter GF, et al. An outbreak of tuberculosis with accelerated progression among persons infected with the human immunodeficiency virus. An analysis using restriction-fragment-length polymorphisms. *N Engl J Med* 1992; **326:** 231–35.

34 Joseph P, Severe P, Ferdinand S, et al. Multidrug-resistant tuberculosis at an HIV testing center in Haiti. *AIDS* 2006; **20:** 415–18.

35 Harries AD, Kamenya A, Namarika D, et al. Delays in diagnosis and treatment of smear-positive tuberculosis and the incidence of tuberculosis in hospital nurses in Blantyre, Malawi. *Trans R Soc Trop Med Hyg* 1997; **91:** 15–17.

36 De Cock KM, Soro B, Coulibaly IM, Lucas SB. Tuberculosis and HIV infection in sub-Saharan Africa. *JAMA* 1992; **268:** 1581–87.

37 WHO. 7th annual report on global tuberculosis control. Geneva: World Health Organization, 2003.

38 Badri M, Wilson D, Wood R. Effect of highly active antiretroviral therapy on incidence of tuberculosis in South Africa: a cohort study. *Lancet* 2002; **359:** 2059–64.

39 Lawn S, Myer L, Bekker L-G, Wood R. Prevalence and incidence of tuberculosis in an antiretroviral treatment (ART) programme in South Africa: risk factors and impact on ART outcomes. Toronto: XVI International AIDS Conference, 2006 (abstr MOPE0175).

40 Jack C, Lalloo U, Karim QA, et al. A pilot study of once-daily antiretroviral therapy integrated with tuberculosis directly observed therapy in a resource-limited setting. *J AIDS* 2004; **36:** 929–34.

41 Dheda K, Lampe FC, Johnson MA, Lipman MC. Outcome of HIV-associated tuberculosis in the era of highly active antiretroviral therapy. *J Infect Dis* 2004; **190:** 1670–76.

42 Farmer P, Furin J, Bayona J, et al. Management of MDR-TB in resource-poor countries. *Int J Tuberc Lung Dis* 1999; **3:** 643–45.

43 O'Brien RJ, Spigelman M. New drugs for tuberculosis: current status and future prospects. *Clin Chest Med* 2005; **26:** 327–40.

44 Lalloo UG, Naidoo R, Ambaram A. Recent advances in the medical and surgical treatment of multi-drug resistant tuberculosis. *Curr Opin Pulm Med* 2006; **12:** 179–85.

45 Barnes L. Dire warning to SA. *The Witness*, Aug 21, 2006. http://www.witness.co.za/default.asp?myAction=sdet&myRef=45847&myCat=news (accessed Oct 2, 2006).

46 WHO. Guidelines for the Programmatic Management of Drug-Resistant Tuberculosis. WHO/HTM/TB/2006.361. Geneva: World Health Organization, 2006.