The emergence, transmission, and control of drug resistant tuberculosis epidemics

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I declare that the work presented in this thesis is my own, submitted for the degree of PhD from Imperial College.



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Abstract

With one third of the world's population infected with tuberculosis, and 8 million new cases of active disease estimated annually, tuberculosis has been declared a global health emergency. Of particular concern has been the finding that drug resistance has developed in every country surveyed, and has reached high levels in a number of settings. Treatment of resistant cases, which require costly and difficult specialized regimens, has added considerable strain to health programs. Crucial questions in the global control of tuberculosis are raised, including where and to what extent will resistance emerge? And, can current control programs adequately manage resistance or are modifications needed? This problem is addressed in this thesis at three levels: the emergence, transmission and control of resistance.

Emergence and transmission

Drug resistance is a man made problem which first emerges by the acquisition of drug resistance mutations during the treatment of a drug sensitive case. This acquired resistance can then be transmitted to new susceptible individuals in the population, who are said to develop primary resistance. A mathematical model for the transmission of tuberculosis was developed on the basis of current understanding of the epidemiology of TB. The structure of the model includes the emergence of acquired and the transmission of primary resistance, in the context of a drug sensitive epidemic. Drug resistance was shown to emerge in a series of linked sub-epidemics following the

start of treatment of a sensitive epidemic. Poor control practices, including a low treatment cure rate and a high acquired resistance rate, allows for the development of a high prevalence of resistance.

Control

In order to consider the impact of different control strategies, the model was expanded to incorporate elements of first and second line control, including case detection, treatment cure rates, drug susceptibility testing, and specialized resistance regimens. In addition, the importance of waiting times to treatment and return of drug susceptibility tests was examined. Case detection and treatment was found to be the most important control measure for reducing disease levels. For controlling resistance, the cure and drug susceptibility testing rates were most important. In high burden countries, control priorities should focus on increasing the case detection and cure rates. In areas where poor treatment practices have already resulted in a high prevalence of resistance, control measures should be first improved before expanding the program. In regions of low incidence, control measures should be improved through detection and treatment of latent infection.

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"There is a dread disease which so prepares its victim, as it were, for death; which so refines it of its grosser aspect, and throws around familiar looks, unearthly indications of the coming change—a dread disease, in which the struggle between soul and body is so gradual, quiet, and solemn, and the result so sure, that day by day, and grain by grain, the mortal part wastes and withers away, so that the spirit grows light and sanguine with its lightening load, and, feeling immortality at hand, deems it but a new term of mortal life; a disease in which death and life are so strangely blended that death takes the glow and hue of life, and life the gaunt and grisly form of death; a disease which medicine never cured, wealth never warded off, or poverty could boast exemption from; which sometimes moves in giant strides, and sometimes at a tardy sluggish pace, but, slow or quick, is ever sure and certain."

-- Charles Dickens, in *Nicholas Nickleby*

Introduction

> "Twenty years ago, tuberculosis, even in its most dangerous form, consumption, was still not considered infectious. Of course, the work of Villemin and the experimental investigations by Cohnheim and Salomonsen had already provided certain clues which suggested that this conception was false. But it was only with the discovery of the tubercle bacillus that the aetiology of tuberculosis was placed on a firm footing, and the conviction gained that this is a parasitic disease, i.e. an infectious, but also avoidable one."
>
> -- Robert Koch – Nobel Lecture ,
> "The Current State of the Struggle against Tuberculosis"
> Nobel Lecture, December 12, 1905

Tuberculosis, an infectious disease caused by *Mycobacterium tuberculosis*, currently causes more deaths than any other single infectious disease. In 2003, the World Health Organization (WHO) reported a global incidence of 8.5 million new cases and 3 million deaths attributable to tuberculosis. One-third of the world's population is infected with *M. tuberculosis*, although only 1 in 10 of those infected will ever progress to active tuberculosis disease. Tuberculosis is particularly concentrated in the developing world, which suffers from 95% of all cases and 98% of deaths. Treatment of the high burden of tuberculosis in the developing world is complicated by poverty and under funded health control programs. (Dye, Scheele et al. 1999; WHO 2000; WHO 2001; Mathers, Stein et al. 2002; WHO 2002; WHO 2003; WHO 2003)

Throughout most of the 19th and 20th centuries, tuberculosis has remained a widespread and deadly disease. Improved living and social conditions, as well as isolation in sanatoria, led to reduced incidence and mortality through the early 1900s.

With the introduction of effective chemotherapy in the 1950s, there was an accelerated decline in both incidence and mortality rates. However, by the early 1990s, there were notable increases in tuberculosis epidemics worldwide. A number of factors are believed to have contributed to the resurgence of tuberculosis, including degradation in program management and funding for tuberculosis and the HIV epidemic. In 1993, the World Health Organization (WHO) declared that tuberculosis posed a global health emergency. (Grzybowski 1991; Styblo 1991; Sudre, ten Dam et al. 1992; Dolin, Raviglione et al. 1994; Kochi 1994; Kochi 1994; Murray and Lopez 1994; Murray, Lopez et al. 1994; Raviglione, Rieder et al. 1994; Raviglione, Snider et al. 1995)

The emergence of drug resistant strains has further complicated control of the resurging epidemic of tuberculosis. In particular, multiple drug resistant (MDR) tuberculosis poses a significant challenge for control programs, as these cases are significantly more difficult and expensive to treat. Epidemics of MDR tuberculosis have been reported, most notably in local outbreaks in New York and the former Soviet Union. (Frieden, Sterling et al. 1993; Viljanen, Vyshnevskiy et al. 1998; Yerokhin, Punga et al. 2001; Munsiff, Bassoff et al. 2002) Recent surveillance has shown that resistance to commonly used anti-tuberculosis drugs have emerged under every treatment program examined. Local hotspots of resistance, with a resistance prevalence of over 30%, were found in several regions. This has alerted concern as to the future course of resistance epidemics, and the potential threat resistance poses to the control of tuberculosis. This has also led to growing concern about the management of resistant cases within program conditions. (WHO/IUATLD 1997; Espinal, Kim et al. 2000; WHO/IUATLD 2000; Davies 2001; Espinal, Laszlo et al. 2001; Raviglione, Gupta et al. 2001)

In this thesis, the concern over resistance within tuberculosis epidemics is addressed at two general levels: emergence and control. First, the conditions under which resistance emerges to high levels are considered. Mathematical models for the emergence and transmission of resistance are analyzed to identify the conditions which allow for the greatest selection for resistance. Questions posed in this section include where does resistance emerge to high levels, and under what situations is there a threat of the emergence of self-sustaining resistant epidemics? When is there cause for concern that resistance levels will continue to increase and interfere with control efforts?

Secondly, the strategies for control of resistance within tuberculosis epidemics are considered. With resistance observed within every region surveyed, what approaches are advisable for management of these resistant cases and prevention of spread of resistance? Finally, how should combined priorities of reducing disease levels and prevention of resistance be incorporated into current programs?

In Chapters 1 to 4 the literature on tuberculosis is reviewed. In Chapter 1 the epidemiology of tuberculosis is presented. The first section of Chapter 1 focuses on the global burden of disease and current practices in surveillance. The second section focuses on resistance, including trends in resistance and observations from recent surveillance of resistance.

In Chapter 2, the molecular biology of tuberculosis is reviewed. This chapter focuses on the properties, lifecycle, transmission, and virulence of *Mycobacterium tuberculosis*.

In Chapter 3, the treatment of tuberculosis is reviewed. The history of the treatment of tuberculosis is followed through the development of effective chemotherapy regimens. The current recommended regimens are explained, as well as

the structure of global control programs, mechanisms for case detection, and tests for diagnosis. In a related chapter placed as Appendix 4, there is a more complete review of the drugs commonly used in current chemotherapy regimens.

In Chapter 4, the contribution of mathematical models to the study of tuberculosis is reviewed. This chapter examines models which have contributed to the understanding of the natural history, the epidemiology, and the intrinsic dynamics of tuberculosis. In the second section, models which examine resistance are presented. The third section includes recent models which are based on the molecular epidemiology of tuberculosis.

In Chapter 5, a mathematical model for the emergence and transmission of resistant tuberculosis is developed. The model construction is explained, the parameters described and sources for parameter estimates given. Appendix 1 provides a more detailed description of the model, including a parameter table, model outline, and the set of differential equations used to define the basic model.

In Chapter 6, the dynamics for the emergence and spread of resistance within TB epidemics are examined using the model presented in the previous chapter. A detailed sensitivity analysis is performed to identify the parameters which most influence model predictions and to characterize the relationship between uncertainty in parameter estimates and model outcome. Appendix 3 provides full details for the sensitivity analyses, and presentation of the simulations not shown within the summary of sensitivity analyses in Chapter 6.

In Chapter 7, the model presented earlier is further developed to include more specific aspects of tuberculosis treatment, to provide a framework to consider the control of tuberculosis and resistance. Appendix 2 provides a more detailed description

of this model, including a parameter table, model outline, and the set of differential equations used to define the basic model.

In Chapter 8, the model is used to examine the impact of elements of first and second line control programs on the dynamics of epidemics and resistance.

In Chapter 9, a more detailed examination of the impact of elements of control on disease and resistance is presented. This chapter focuses on the priorities for control in representative settings which differ by disease burden, resistance prevalence, and level of control.

Chapter 10 provides a final summary and discussion of the entire thesis, along with brief consideration of implications of this research and future studies.

1. Chapter 1

Epidemiology of tuberculosis

"But let me add a warning: do not get snobbish. I remember I used to sort of despise the untutored, ignorant folk who did not have or had not had TB. I looked down upon such unfortunates as an inexperienced, inferior lot, who after all, couldn't know much about life or anything else. They simply didn't belong, thought I with a superior sniff--until one day a friend, an eminent TB specialist, sensing my attitude, maliciously told me that truth--that autopsies reveal the democratic fact that nearly everyone has had it! Which leaves us only one point of superiority to brag about: We know it and the rest of them don't."

-- Eugene O'Neill, in a message to patients with tuberculosis

1. CHAPTER I

EPIDEMIOLOGY OF TUBERCULOSIS

I. GOALS

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III. GLOBAL BURDEN

III. A. MORTALITY
III. B. INCIDENCE RATES
III. C. CASE NOTIFICATIONS

IV. HIGH BURDEN COUNTRIES

IV. A. TRENDS BY REGION

V. MEASURES OF TB BURDEN

V. A. ESTIMATES OF THE PROPORTION INFECTED AND ARI
V. B. CURRENT SURVEILLANCE
V. C. MORTALITY

VI. DOTS

VII. RESISTANCE

VHI. RESISTANT STRAINS TRANSMISSION

IX. SUMMARY

I. Goals

I. A. To review the epidemiology of tuberculosis.

I. B. To review the methods for measuring and surveillance of tuberculosis disease burden.

I. C. To review trends in resistance to tuberculosis drugs.

II. Abstract

Tuberculosis, an infectious disease caused by *Mycobacterium tuberculosis*, is a leading cause of death globally. One third of the world's population is infected with *M. tuberculosis*, although only about one in ten of infected individuals will experience active disease in their lifetime. The annual global incidence of tuberculosis is estimated at 8.5 million new cases, and of these 3.7 million smear positive cases. 3 million deaths are attributed to tuberculosis annually, making tuberculosis the 8th leading cause of death globally, and second to HIV/AIDS amongst infectious diseases. The burden of disease is disproportionately found in developing countries, with over 90% of cases located in 22 designated high burden countries (HBC). The common measures for surveillance of tuberculosis include incidence and prevalence rates, mortality rates, and prevalence of resistance.

Resistance to antituberculosis drugs has been observed since the introduction of chemotherapeutic treatment in the 1960s. Improved regimens, including use of multiple drugs and the introduction of the more effective drug rifampicin, allowed for reduced rates of resistance amongst treated cases (for review of commonly used drugs see Appendix 4). With the resurgence in tuberculosis epidemic in the early 1990s, there were also noticeable increases in resistance prevalence in a number of locations.

Most notably, outbreaks of MDR in New York City and increases in overall resistance in Eastern Europe alerted concerns over treatment practices. In 1993, the World Health Organization (WHO) began surveillance for tuberculosis resistance amongst member countries. Surveillance showed the existence of hotspots of resistance, with six regions found to have over 30% prevalence of resistance to at least one drug and over 3% for MDR.

III. Global burden

Tuberculosis, an infectious disease caused by *Mycobacterium tuberculosis*, annually causes more deaths than any other single pathogen. An estimated 2 billion individuals, one third of the world's population, are infected with *M. tuberculosis*. While many are infected with the bacteria, only about 10% of infected individuals ever develop active TB disease, while the rest remain latently infected. In 2002, WHO estimated a global incidence of 8.5 million new cases of active TB, with 3.7 million smear positive cases. (Dye, Scheele et al. 1999; WHO 2000; WHO 2003)

III. A. Mortality

An estimated 3 million deaths annually are attributable to TB, accounting for 3% of all deaths worldwide. Tuberculosis is the 8th leading cause of death globally. (Table 1-1, (Mathers, Stein et al. 2002)) Amongst infectious diseases, TB is the second largest cause of deaths, after HIV/AIDS. (Table 1-2, (Mathers, Stein et al. 2002)) Together, infectious and parasitic diseases accounted for 19.3% of all global deaths in 2001. Amongst communicable diseases (this classification includes infectious and parasitic diseases), diseases HIV/AIDS was the leading cause of death, accounting for 5.1% of global deaths. This is followed by diarrhoeal diseases, accounting for 3.5% of global deaths. Tuberculosis is the third leading cause of

deaths amongst communicable diseases, accounting for 2.9% of global deaths. With over 90% of TB cases found in developing countries, this affect is disproportionately seen in the developed world. In developing countries, tuberculosis is again the 8th leading cause of death, accounting for 5% of deaths. In comparison, in developed countries, tuberculosis is not found amongst the top ten causes of death. (Mathers, Stein et al. 2002)

	Ten leading causes of deaths, global estimates 2000	% of total deaths
1	Ischaemic heart disease	12.6%
2	Lower respiratory infections	11.1%
3	Cerebrovascular disease	9.6%
4	Chronic obstructive pulmonary disease	4.7%
5	HIV/AIDS	4.6%
6	Perinatal conditions	4.5%
7	Diarrhoeal diseases	3.6%
8	Tuberculosis	2.9%
9	Road traffic accidents	2.2%
10	Trachea, bronchus, lung cancers	2.1%

Table 1-1. ***Ten leading causes of deaths, from 2000 global estimates.***

The top ten leading causes of deaths amongst all countries represented in the survey. Amongst infectious diseases, HIV/AIDS accounted for 4.6% of all deaths, and TB for 2.9%. Source: Table reproduced from (Mathers, Stein et al. 2002).

	Number of deaths	% of total global deaths
All Infectious and parasitic diseases	10,937,452	19.3%
HIV/AIDS	2,865,804	5.1%

Diarrhoeal diseases	2,001,193	3.5%
Tuberculosis	1,644,381	2.9%
Childhood-cluster diseases	1,318,151	2.3%
Malaria	1,123,764	2.0%

Table 1-2. ***Total global mortality rates for infectious and parasitic diseases.*** Together, infectious and parasitic diseases accounted for 19.3% of all global deaths in 2001. Amongst communicable diseases, HIV/AIDS was the leading cause of death, accounting for 5.1% of global deaths. This is followed by diarrhoeal diseases, accounting for 3.5% of global deaths. Tuberculosis is the third leading cause of deaths amongst communicable diseases, accounting for 2.9% of global deaths. Estimates for deaths attributable to communicable diseases as reported in the World Health Report 2002, which provides estimates for mortality, incidence, prevalence, YLL, YLD and DALYs by sex, age and cause in 6 WHO regions in 2001. Source: (WHO 2002).

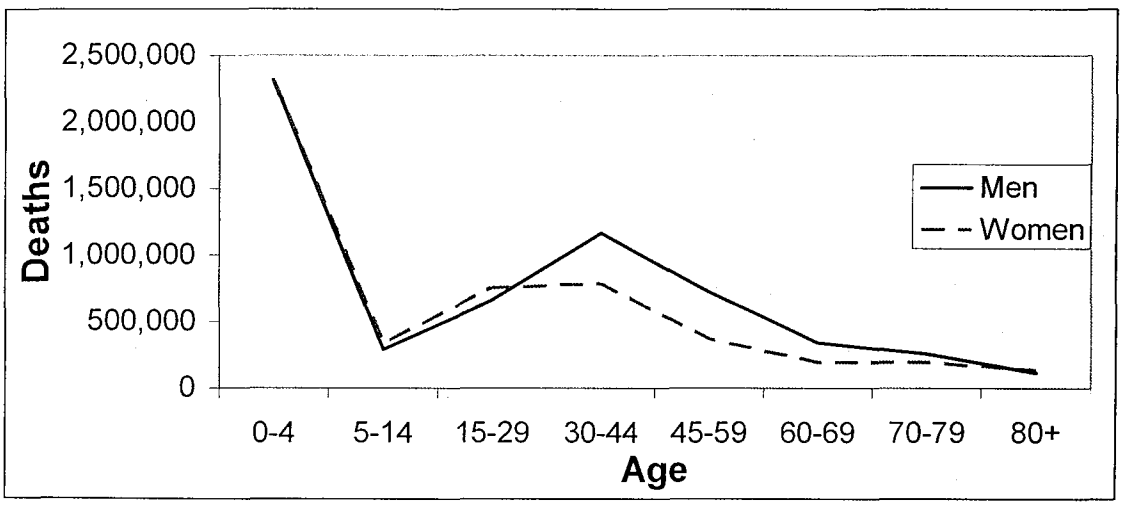


Figure 1-1. ***The number of deaths attributable to tuberculosis according to gender and age in 2001.*** Estimates for deaths attributable to communicable diseases as reported in the World Health Report 2002. The highest death rates due to TB were found in children aged 0-4, followed by those in the age group 30-44. The death rate was slightly higher in men than in women for ages 30-80. Source: (WHO 2002).

III. B. Incidence rates

Over 80% of TB cases occur in 22 countries designated by the WHO as High Burden Countries (HBC). The average annual incidence rate in the HBCs is 80 cases per 100,000, ranging from 30 to 260 cases per 100,000. (Netto, Dye et al. 1999; WHO 2003) Regions with less than 10 cases per 100,000 are considered low

incidence, while areas with less than 1 case per 100,000 are entering the elimination phase, and below 0.1 cases per 100,000 TB are considered eliminated. (Clancy, Rieder et al. 1991; CDC 1998; Broekmans, Migliori et al. 2002)

Studies have suggested that an annual risk of infection of 1% corresponds to an annual incidence rate of approximately 50-100 cases per 100,000. With this assumption, we consider high incidence rates, as found in the HBCs, ranging from 50 - 250 cases per 100,000 may correspond to an ARI of approximately 0.5 – 2.5%. Other studies have used similar estimates, such as an ARI of 1.5-2.5% in Africa, 0.5-1.5% in Americas, and 1-2.25% in Southeast Asia and Western Pacific. (Sudre, ten Dam et al. 1992) We consider low incidence rates, of below 10 cases per 100,000, to correspond to an epidemic with an ARI of less than 0.1%. We use these estimates as rough guidelines for considering the timescale of epidemics.

III. C. Case notifications

In 2003, 183 countries contributed reports of case notifications to the WHO survey. 3.8 million new cases of TB were notified, with an notification rate of 62 cases per 100,000 population. Of these, 1.6 million (42%) were smear positive. In comparison, estimates suggest that there are 8.5 million new cases, with 3.7 million smear positive, annually. The reported case notifications in 2003 represents only 45% of the estimated new cases and 43% of the smear positive cases. (Dye, Watt et al. 2002; WHO 2003)

The overall global notification rate of TB cases has remained nearly constant since WHO surveillance records began in 1980. Meanwhile, the number of cases enrolled in DOTS programs has increased much faster. The average global notification rate has remained nearly constant since 1980, fluctuating between 50 – 70 sputum smear positive cases per 100,000. (Figure 1-2, (WHO 2003)) Recent

estimates show detection of about 30-40% of global incidence. The WHO has set a target for detection of 70% of incidence. (Dye, Watt et al. 2002; WHO 2003)

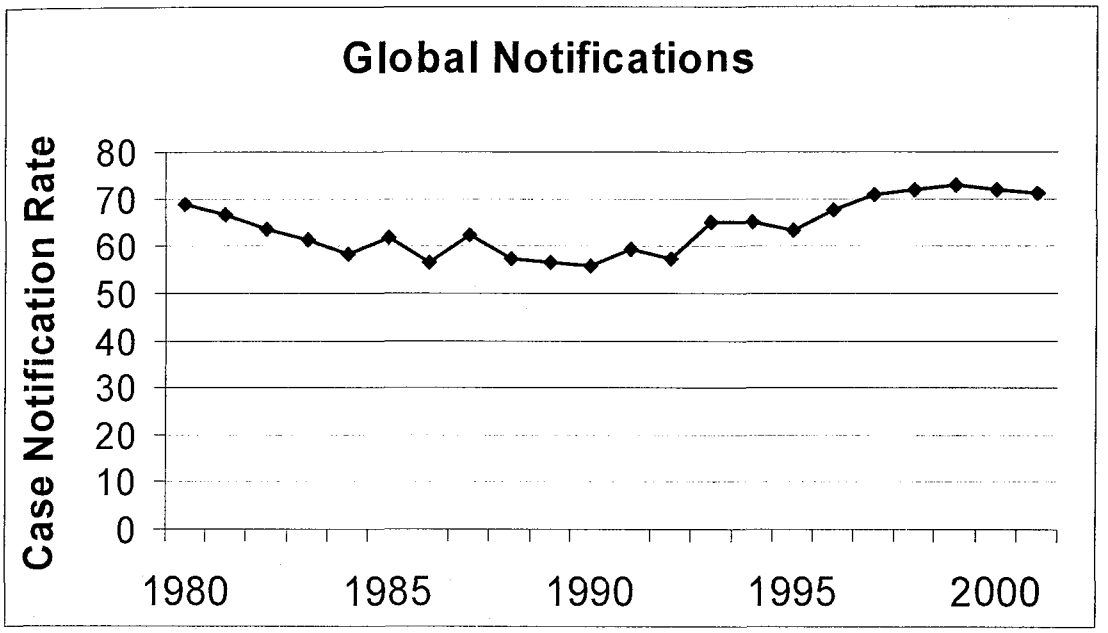


Figure 1-2. Global notification rate of smear positive cases. Source for case notification rates: (WHO 2003).

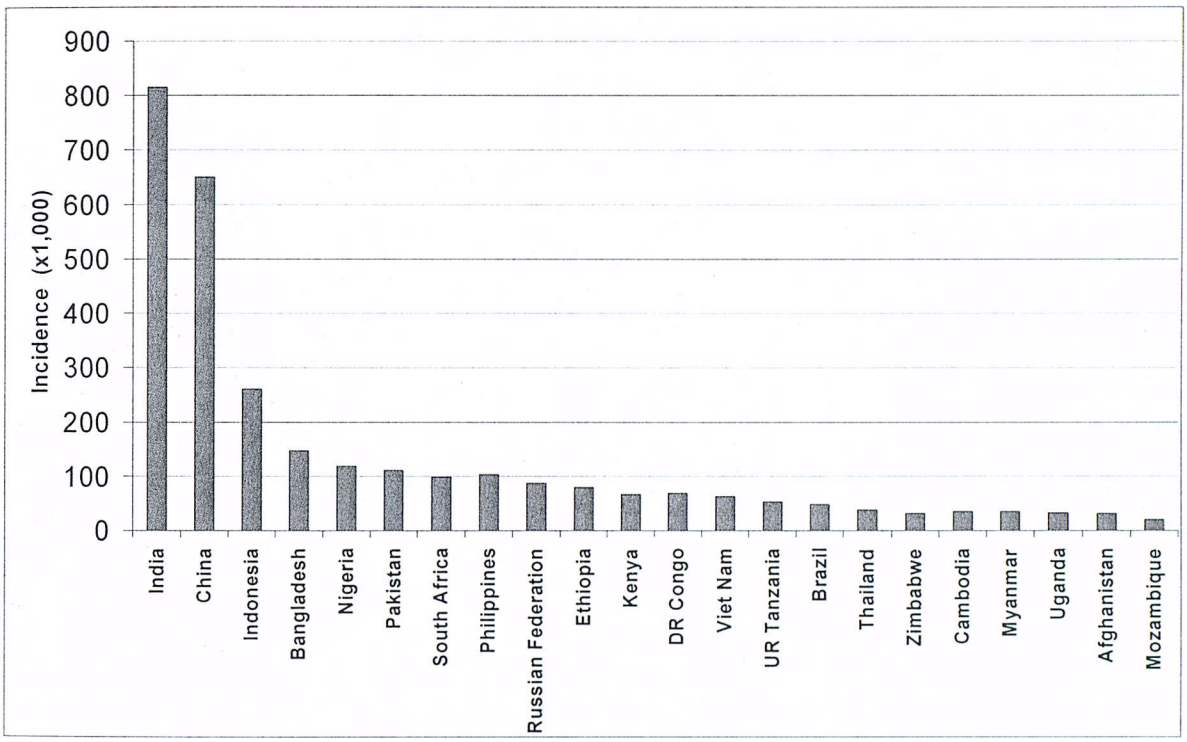
IV. High burden countries

Over 80% of new TB cases are found in 22 high burden countries (HBC). The regions with the highest incidence rates include Southeast Asia, sub-Saharan Africa, and eastern Europe. 37% of the global incidence is found in the South-East Asia region, 21% in the African region, and 22% in the Western Pacific region. Together, these three regions account for 80% of notified cases. (WHO 2003)

The number of new cases found in the high burden countries in 2002, as reported by the WHO, is shown in Figure 1-3A. The highest overall number of cases are found in India (815,000 cases), China (651,000 cases), Indonesia (261,000 cases), Bangladesh (147,000 cases), and Nigeria (119,000 cases). The standardized incidence rates per 100,000 population are shown in Figure 1-3B. The standardized incidence

rates range from 28 cases per 100,000 in Brazil to 261 cases per 100,000 in Cambodia. The top five highest standardized incidence rates are found in Cambodia (261 cases per 100,000), Zimbabwe (252 cases per 100,000), South Africa (226 cases per 100,000), Kenya (213 cases per 100,000), and Tanzania (146 cases per 100,000). (Figure 1-3)

A.



B.

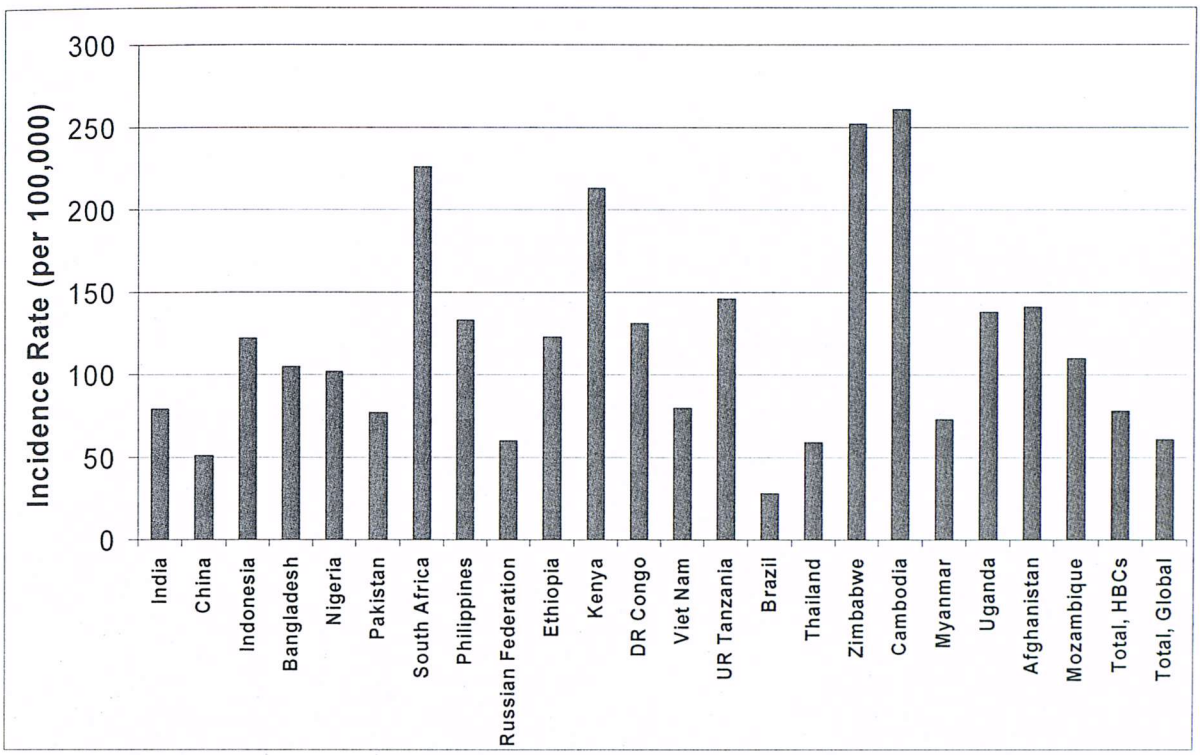


Figure 1-3. ***Incidence in the 22 high burden countries.*** The disease burden found in the 22 high burden countries is shown in (A) as the estimated thousands of cases in (B) as the estimated incidence rates per 100,000 population. Countries are listed from left to right in order of highest number of new cases. Source for incidence and incidence rates: (WHO 2003).

IV. A. Trends by region

Recent trends in tuberculosis epidemics vary distinctly by region. In the developed market economies, over the past twenty five years, there has been a marked reduction in disease levels. In comparison, in many developing countries, slower implementation of control programs and limited resources have allowed for a continued increase. In regions with high prevalence of HIV, there has been a notable increase in incidence rates, fueled by the HIV epidemic.

Shown in Figure 1-4 are the trends in case notifications by WHO regions. Case notifications have been recorded in these regions since 1980. Amongst these regions, notification rates have been increasingly notably only in Africa. In

particular, there have been significant increases in regions of Africa with a high prevalence of HIV. Within the other regions, the average notification rates have stayed fairly constant in recent years. The lowest notification rates are found in the regions of the Americas and Europe. Over the past five years, there has been a slight decrease in notification rates in the Americas, and a slight increase in Europe. (Figure 1-4)

In Figure 1-5, the smear positive notification rates, as recorded by WHO surveillance since 1993, are shown for each region. While the trends for each region are similar to that seen in Figure 1-4 with the total notification rates, several additional observations can be made here. First, the increase in case notifications in Africa in the past decade has come disproportionately from smear positive cases. While in 1993 smear positive cases accounted for about one fourth of total notifications, by 2001 smear positive cases accounted for nearly one half of total notifications. Secondly, there has been an increase in smear positive notifications in the Southeast Asia region, which was not discernable from trends in the total notifications. Again, the increase in case notifications appears to be disproportionately amongst smear positive cases. Similarly, there were slight increases in smear positive notification rates in both the Western Pacific and Eastern Mediterranean regions which were not discernable from total notification rates. Third, while there appears to be a slight increase in total notifications in Europe over the past five years, the smear positive notification rate has stayed nearly constant at 10 cases per 100,000. The frequency of smear positive cases in the Europe region has declined from one in four (in 1993) to one five cases (in 2001). (Figure 1-5)

As can be seen in Figure 1-5, the region with the lowest smear positive notification rates over the past decade is Europe, with a smear positive notification

rate that has stayed nearly constant at 10 cases per 100,000. The regions of the Western Pacific, the Eastern Mediterranean, and the Americas all have maintained smear positive notification rates near or below 20 cases per 100,000 since 1993. In the Southeast Asia region, smear positive notification rates have increased from 20 in 1993 to nearly 40 in 2001. The greatest increase was seen in the Africa region, with an increase from 20 in 1993 to nearly 60 smear positive cases per 100,000 in 2001. (Figure 1-5)

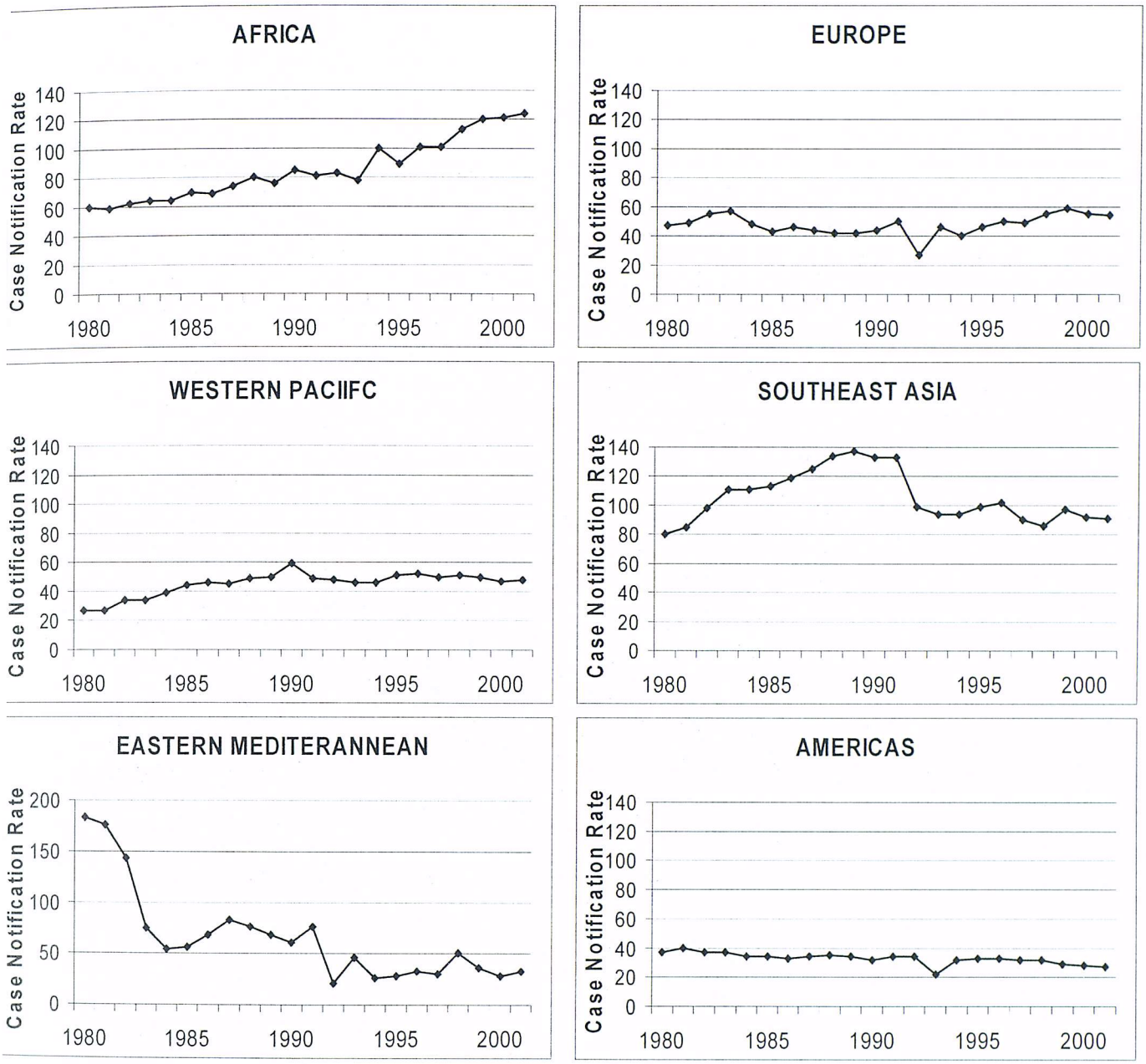


Figure 1-4. ***Regional trends in the notification rates by region since 1980.*** The case notification rates are given as new cases per 100,000 population in each of these six regions: Africa, Southeast Asia, Western Pacific, Europe, Eastern Mediterranean, and Americas. Source for the case notification rates is from (WHO 2003).

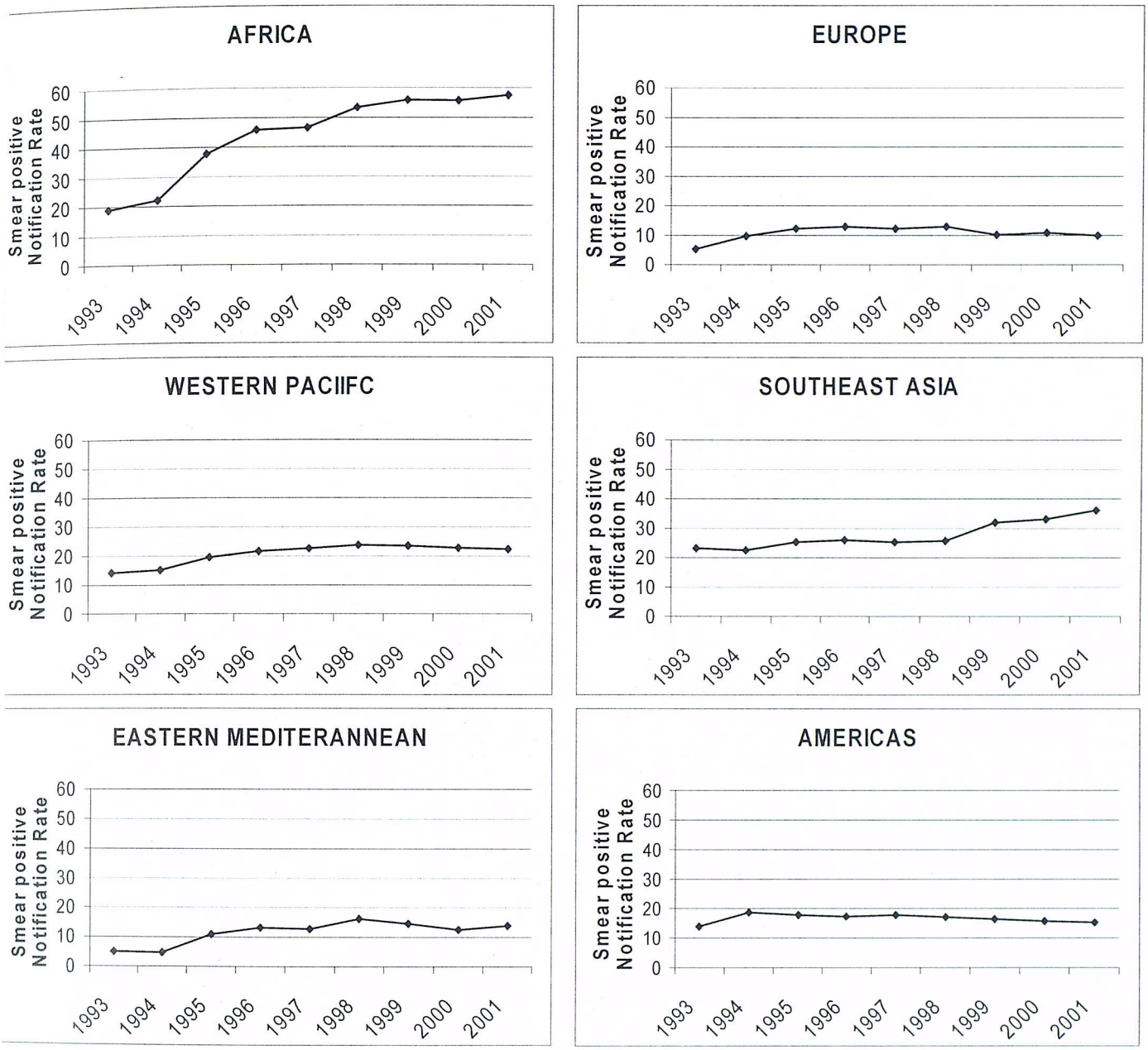
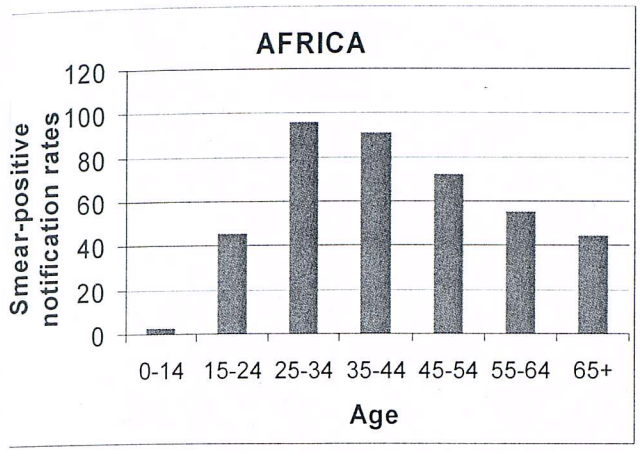
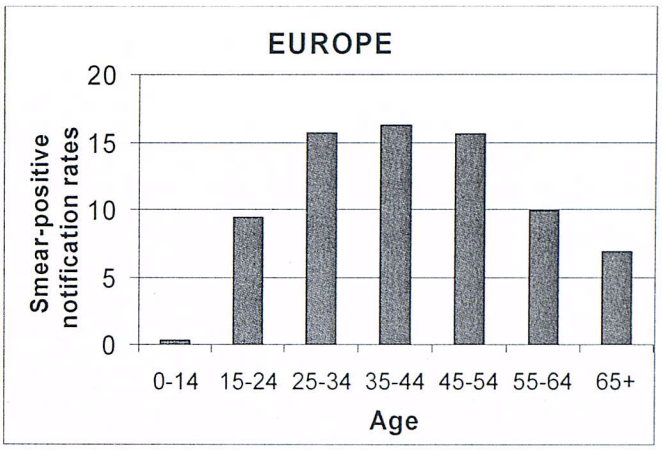
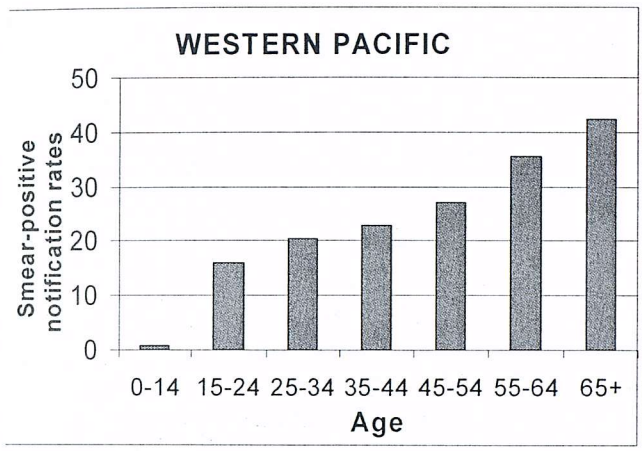
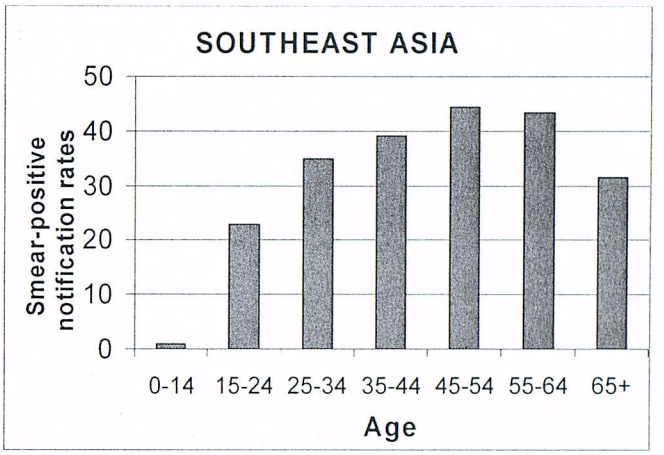


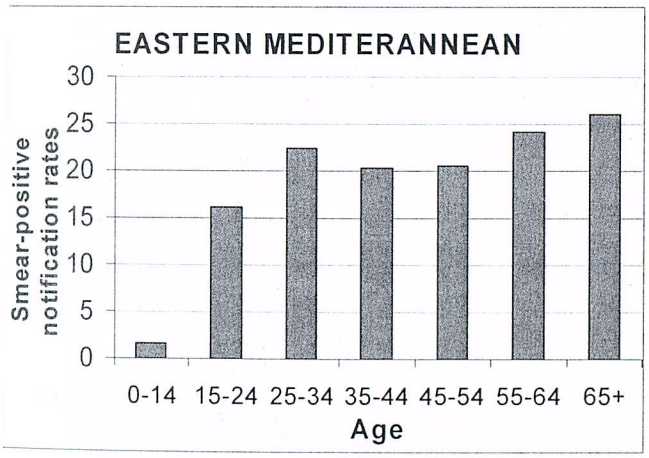
Figure 1-5. ***Regional trends in smear positive notification rates since 1993.***
The smear positive notification rates are given as new cases per 100,000 population in each of these six regions: Africa, Southeast Asia, Western Pacific, Europe, Eastern Mediterranean, and Americas. Source for the case notification rates is from (WHO 2003).











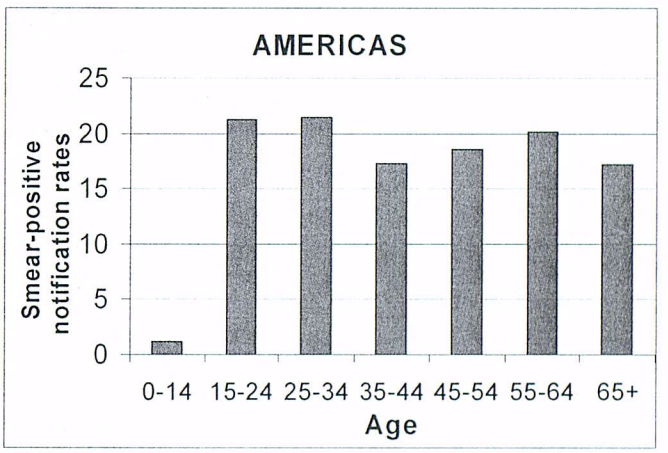


Figure 1-6. ***Smear positive notification rates by region and age.*** The smear positive notification rates are given as new cases per 100,000 population in each of these six regions: Africa, Southeast Asia, Western Pacific, Europe, Eastern Mediterranean, and Americas. Note that different scales are used for the notification rates in each figure, to allow for comparison of the relative incidence by age within each region. Source for notification rates is (WHO 2003).

V. Measures of TB burden

Tuberculosis burden can be measured in a number of different ways, including mortality rates, case numbers, infection prevalence, incidence rate, and annual risk of infection. The introduction of chemotherapy in the 1960s has altered the relative use of these measurements in surveillance. In the prechemotherapy era, a good measure of TB burden in a region was obtained through mortality rates. Under no treatment, the TB mortality rate was estimated at approximately 50% of the incidence rate. However, with improvements to treatment, the mortality rates have been significantly reduced, and this is no longer a reliable indicator of disease burden. (Enarson and Murray 1996; Davies 1998)

Another measure for TB epidemics is simply case numbers in a population. In the prechemotherapy era, there was an estimated 2:1 ratio of prevalence to incidence. This translated into an average case duration of infectiousness of 2 years. However, treatment has reduced the average case duration of infectiousness and disrupted this ratio. Estimating case numbers now provides a good measure of the burden and density of disease at a particular moment in time. (Enarson and Murray 1996)

V. A. Estimates of the proportion infected and ARI

The prevalence of TB infection in a population can be estimated through surveys using the tuberculin skin test. However, use of the skin test to estimate prevalence of infection is complicated by test specificity. In particular, the test may show positive for detection of other mycobacteria, particularly in regions with high environmental mycobacteria, or due to BCG vaccination. Once the prevalence of infection has been estimated, from serial skin test surveys within a population, the annual risk of infection may be estimated. The annual risk of infection represents an

estimate of the probability of infection. Again, difficulties with this include those inherent to the skin test, including infection with non-tuberculosis mycobacteria.

The annual risk of infection was first estimated with models developed by Styblo, using data from both developed and developing nations. In these models, a relationship between the incidence of smear positive cases and the risk of acquiring new infection is calculated. This study estimated that there is an annual risk of infection of 1% for each 49 smear positive cases (also referred to as the 1:50 rule). This has been held as the 1:50 rule for estimating incidence. However, the introduction of treatment has disrupted this ratio, which is expected to increase as incidence decreases under good treatment. (Styblo 1985)

V. B. Current surveillance

The most common measure today for TB burden has become incidence of disease. The incidence rate is typically measured as a rate of new cases per population of 100,000, in order to allow for standardized comparison. In addition, measures of disease prevalence provide a snapshot of the density of disease at a particular moment. The standardized case rate is reported in WHO surveillance studies as the number of cases present per 100,000 population. Understanding the trends in regional TB control programs is also important in understanding the state of a particular TB epidemic. Measures reflecting the level of TB control include the rates of detection and treatment success (including completion and cure). In addition, the proportion cases reporting under DOTS programs, reflects on the program, with estimated cure rates under both DOTS often much higher than those under non-DOTS programs. (WHO 2003)

V. C. Mortality

In the prechemotherapy era, mortality rates were a good indicator of the TB epidemic. There was a nearly constant 1:2 relationship between incidence and mortality. However, the introduction of effective treatment significantly reduced the death rates amongst TB cases, and other measures provided better indicators of the disease levels. Even before the introduction of chemotherapy, the mortality rates showed significant declines in the early 1900s, correlated to improvements in overall health care and standards of living. In the early 1900s, there was a decline in mortality on average of 4% per year. From 1945 to 1960, there was continued decrease in mortality rates in industrialized countries, followed by a stabilization coinciding with the introduction of effective chemotherapy including use of streptomycin and isoniazid in treatment of cases. (Davies 1998) (Enarson and Roullin 1998)

VI. DOTS

The WHO strategy for TB control is known as DOTS (Directly Observed Treatment, Short Course). The DOTS strategy is defined by five central elements, which include, but reach far beyond recommendations for standardized treatment regimens. The five central elements defining DOTS programs are: government commitment, case detection, short course chemotherapy, regular supply of all essential anti-tuberculosis drugs, and a standard reporting system for notifications and treatment results. The treatment regimens are standardized, with regimens for new cases consisting of at least four drugs. The short course chemotherapy (SCC) regimens for new cases last from six to nine months, with direct observation for at least the first two months of treatment. Case detection is through self-reporting and

diagnosis through sputum smear microscopy. (Weis, Slocum et al. 1994; Wise 1998; WHO 1999; WHO 2003)

Since 1995, over 10 million cases have been diagnosed and treated under DOTS. As of 2002, 155 countries implement DOTS programs to some extent, with 61% of the world's population living in areas with access to these programs. In 2002, 2.4 million new cases (1.2 million smear positive cases) were notified under DOTS program. About 29% of total estimated incidence and 32% of estimated smear positive incidence are notified under DOTS. (WHO 2003)

DOTS programs have consistently maintained higher cure rates, achieving cure rates as high as 95%. The global average for treatment cure under DOTS is 78%, compared to 45% reported under non-DOTS programs. DOTS programs maintain higher standards of treatment, increasing cure rates and reducing mortality as well as resistance. (Wise 1998) (Raviglione, Dye et al. 1997; Dye, Watt et al. 2002) Under non-DOTS programs, in comparison, there are far more variable outcomes. Non-DOTS programs show lower rates of treatment success, unreliable outcomes, higher rates of chronic cases, and higher rates of acquired resistance. However, comparing non-DOTS programs directly is difficult because most lack standardization of methods and variable reliability and recording of case outcomes. (Raviglione, Dye et al. 1997; WHO 1999; WHO 2003)

DOTS has been ranked by the World Bank as one of the most cost effective health interventions. (Wise 1998) In many of the HBCs, DOTS offers a low cost alternative within health services with limited funding, political support, and technical expertise. (WHO 1999; Wilton, Smith et al. 2001; WHO 2003) Maintaining this low cost is important in order to allow viable implementation within the developing world, where most cases are found.

VII. Resistance

The emergence of drug resistant strains has further complicated tuberculosis control. Epidemics of multiple drug resistant (MDR) tuberculosis have been reported, including widely publicized outbreaks in New York and the former Soviet Union. MDR tuberculosis poses a significant challenge for treatment programs, as this form of tuberculosis is significantly more difficult and expensive to treat. (Eltringham and Drobniewski 1998; Bleed, Dye et al. 2000; Dye, Fengzeng et al. 2000; Espinal, Kim et al. 2000; Espinal, Laszlo et al. 2001; Raviglione, Gupta et al. 2001)

Clinical resistance is defined as a case that emits resistant mycobacteria in their sputum. Mycobacterial confirmation of resistance, through culture or other drug susceptibility tests, is required to designate a case as resistant. (See Chapter 2 for more on resistance testing) However, resistance may be suspected amongst any cases which fail first line treatment. Under many programs, in particular in developing countries, resources for drug susceptibility testing of all cases are not available. Under these circumstances, cases which fail one or more courses of first line treatment, should be suspected of having resistance, and may be placed on retreatment regimens.

Primary resistance is defined as a new case which has been infected with a resistant strain. By definition, cases with primary resistance have received no previous treatment. In contrast, cases with acquired resistance have developed resistance during a course of treatment, and have received at least four weeks of previous treatment. However, because it has proved difficult to correctly identify cases as having primary or acquired resistance, the WHO has changed classifications to differentiate between resistant cases based on previous treatment. Cases with "initial" resistance are defined simply as those which present with resistance, without

reference to treatment history. Resistant cases with previous treatment are defined as those that have received at least one month of previous treatment and do not respond to first line regimens.

Cases continuing to excrete bacilli in sputum after 5 months of treatment are designated as treatment failures, and may be placed on retreatment regimens. DOTS retreatment regimens usually consist of five drugs, including use of isoniazid, rifampicin, and ethionamide throughout, and pyrazinamide and ethambutol in the initial phase. Under proper administration, most of the cases which remain infectious after 5 months of a retreatment regimen are resistant to at least one of the first line drugs. Cases which fail a retreatment regimen are designated as chronic cases, having received at least two courses of therapy. Chronic cases show high rates of acquired resistance and often MDR. (WHO 1997; Shimao 1998; WHO 1999; WHO 2003)

Resistance has been observed since the introduction of chemotherapy for tuberculosis. In the early regimens, treatment with only one drug, isoniazid or related compounds, allowed for high rates of resistance. (Buck 1952; Hobby 1952; Steenken 1952; Szybalski 1952; Barnett 1953) Studies also observed high rates of resistance when patients were treated with either streptomycin or PAS alone. (Mitchison 1950) Analysis of these early studies showed that these regimens could improve patient symptoms and prolong life, but cases remained infectious and developed resistance to the drugs used. (Mitchison 1969) Including a second drug in the treatment regimen was shown to reduce the observed rates of resistance to either drug, when compared to monotherapy. (Tripathy, Menon et al. 1969) With the introduction of rifampicin in the early 1970s, the treatment of TB improved, allowing regimens to be reduced to 6 months while achieving higher cure rates. Use of rifampicin during the initial two months of treatment appeared to give needed bactericidal and sterilizing activity to

clear a large proportion of the mycobacteria during the early phases of treatment. (Dickinson and Mitchison 1970; Mitchison 1970; Dickinson and Mitchison 1981) With the use of short course chemotherapy regimens, containing four or five drugs including rifampicin, there was an improvement not only in treatment cure rates, but also in a reduction in resistance. Including rifampicin in the early phase of a treatment regimen significantly reduced the rates of resistance to any of the drugs used. (Fox and Mitchison 1975; Dickinson and Mitchison 1976; Fox and Mitchison 1976) (See Appendix 4 for more details on drug action of rifampicin and other drugs commonly used within chemotherapy regimenss).

Early studies also examined the treatment of resistant cases. Cases with initial resistance showed higher rates of treatment failure and relapse. Cases with resistance to a single drug, isoniazid or streptomycin, could be cured with extended regimens containing four drugs including rifampicin. However, treatment of cases with resistance to both isoniazid and rifampicin were far more difficult; over 70% of these cases failed treatment or relapsed. (Mitchison and Nunn 1986)

With the resurgence of TB epidemics in the early 1990s, the resistance problem has also grown. Epidemics of resistance were reported in a number of areas. The highest prevalences of MDR were found in Nepal (48.0%), Gujarat, India (33.8%), New York City, (30.1%), Bolivia (15.3%), and Korea (14.5%). (Cohn, Bustreo et al. 1997)

The United States also showed considerable increases in overall TB incidence from 1985 to 1992. In areas of highest burden, the epidemic appears to have been fueled by poor control and treatment practices, and within high risk groups, including immigrants and HIV coinfected. (McCray E 1997) New York City was a prominent high burden area, in which the incidence of TB nearly doubled during the early 1990s.

Resistance and MDR outbreaks contributed to the NYC epidemic, Amongst all cases tested by culture in NYC in 1991, over 30% showed resistance, and 19% were resistant to both isoniazid and rifampicin (MDR). Amongst cases which had received prior treatment, 44% had resistance to at least one drug, and 30% were resistant to both isoniazid and rifampicin. Cases coinfected with HIV and injection drug users were more likely to show resistance. (Frieden, Sterling et al. 1993) A high proportion of new resistant and MDR cases were attributable to ongoing transmission, representing primary resistance. The overall level of resistance to any drug amongst new cases increased from 10% in 1982 to 23% in 1991. RFLP analysis of strains showed clustering of a high proportion of MDR cases, providing additional support for high rates of recent MDR transmission. (Alland, Kalkut et al. 1994; Friedman, Stoeckle et al. 1995; Bifani PJ 1996; Munsiff, Bassoff et al. 2002)

Eastern Europe has also suffered significant increases in resistance levels over the past decade. In the Russian Federation, the rates of morbidity, mortality, and relapse for tuberculosis have increased along with incidence since 1991. These changes are correlated to increases in drug resistance. (Viljanen, Vyshnevskiy et al. 1998; Davies 2001; Yerokhin, Punga et al. 2001)

In order to quantify the extent of the resistance problem, the WHO and the International Union against Tuberculosis and Lung Disease (IUATLD) started the Global Project on Anti-tuberculosis Drug Resistance Surveillance in 1994. This project coordinated a global resistance surveillance effort within 58 regions from 1994 to 2000, presented in two WHO reports. (WHO/IUATLD 1997; WHO/IUATLD 2000) Drug resistance was found in every region surveyed. There was a wide range in resistance reported; the resistance prevalence in new cases ranged from 1.7-37% (median 10.7%) in countries surveyed. The prevalence of resistance to

a single drug ranged from 2.9-40.8%. Prevalence of MDR, defined as resistance to at least isoniazid and rifampicin, ranged from 0 -18%. Six regions were found to be hotspots of MDR, with greater than 3% MDR prevalence. (Figure 1-7) (WHO/IUATLD 1997; WHO/IUATLD 2000)

The resistance prevalence within a region was found correlated to a number of factors, in particular treatment patterns. Higher resistance rates were found in regions with poor program conditions. The resistance prevalence was negatively correlated to the proportion of cases notified under DOTS programs. Higher resistance prevalence was also correlated to a greater proportion of previously treated cases registered, another measure of treatment quality. In addition, higher resistance rates were correlated to the time since the introduction of rifampicin and since National Treatment Program (NTP) formation. (WHO/IUATLD 2000)

Using data from several of the WHO studies, we examined the relationship between resistance prevalence and treatment. Shown in Figure 1-8 is the prevalence of single (DR) and multiple drug resistance (MDR) by the treatment and cure rates found in that region. The prevalence of both DR and MDR are seen to be inversely proportional to the cure rate. The prevalence of resistance also decreases with increasing notification rates; however the strength of this correlation is less. (Figure 1-8) In order to further explore the nature of the relationship between resistance and treatment rates, the correlation between the resistance prevalence and notification was examined stratified by program cure rates. When the program cure rates were high, increasing the notification rates was correlated with reductions in the prevalence of both DR and MDR. However, at lower program cure rates, increasing the notification rates had more variable outcomes, and could increase the prevalence of DR and MDR. (Figure 1-9)

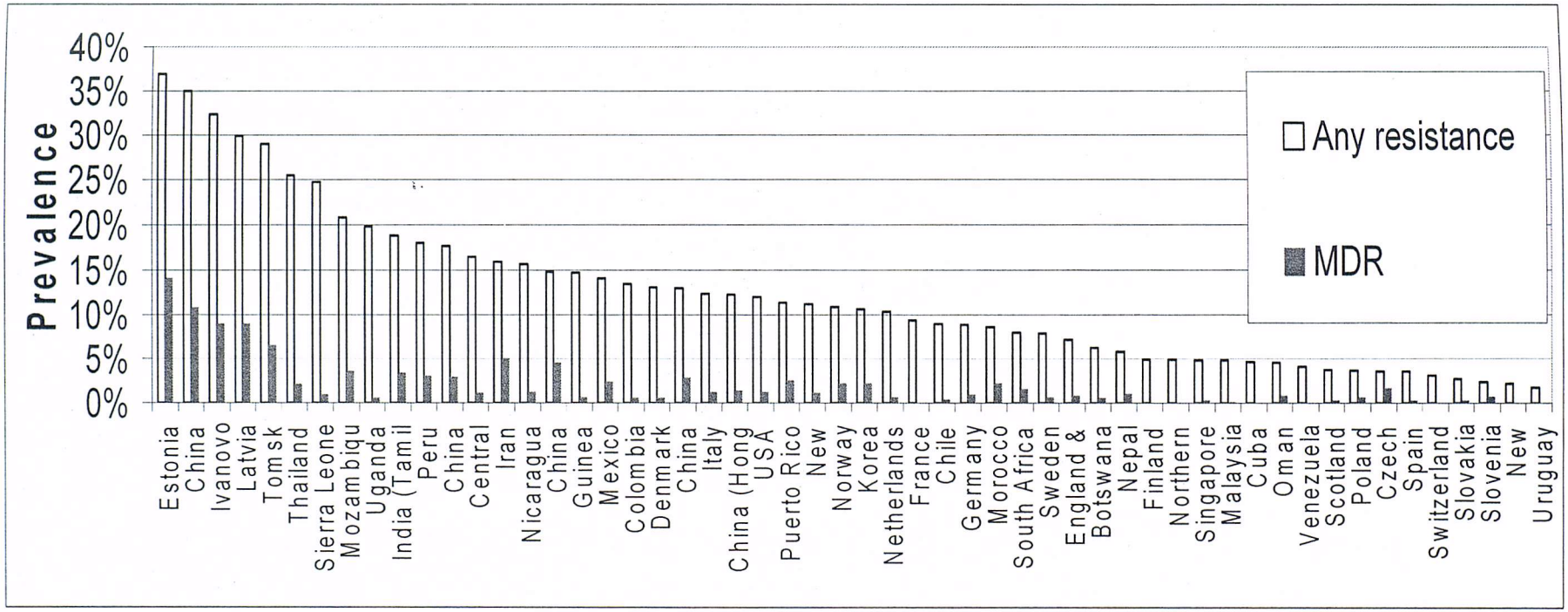


Figure 1-7. ***Prevalence of resistance within all regions surveyed in the WHO/IUATLD 2000 report.*** The prevalence of any resistance and MDR is given as a proportion of all TB cases found in each region. Regions are listed from left to right in order of resistance prevalence. Source: prevalence of resistance as reported by the WHO/IUATLD Global Project on Anti-tuberculosis drug resistance surveillance, from (WHO/IUATLD 1997; WHO/IUATLD 2000).

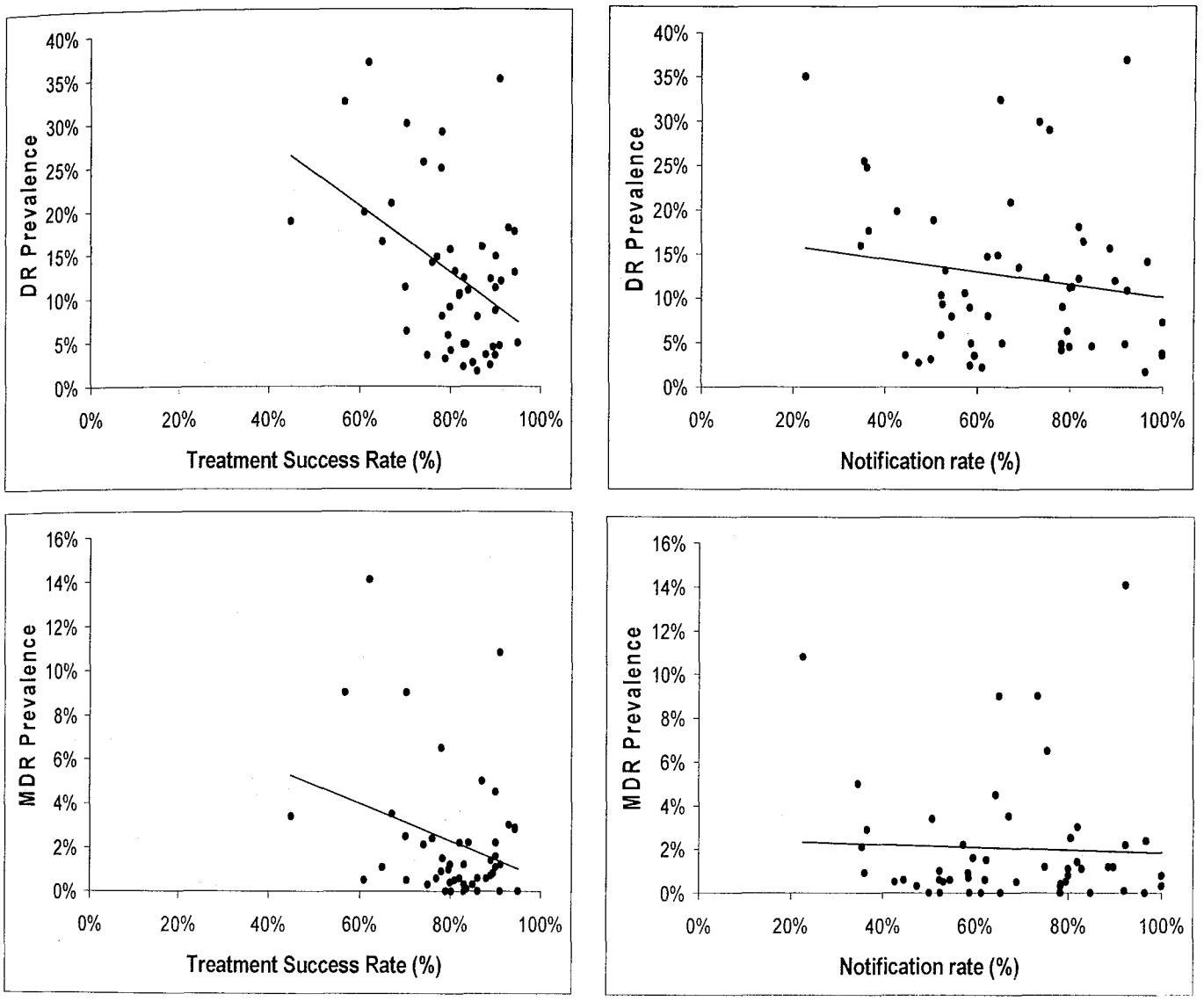


Figure 1-8. *Treatment and prevalence of resistance: higher rates of treatment notification and success are correlated to lower prevalence of resistance.* For each region which reported a resistance prevalence within (WHO/IUATLD 2000), the corresponding treatment success and notification rates for each region were found from (WHO 2003). The prevalence of any resistance (DR Prevalence) and MDR (MDR Prevalence) is shown relative to either the treatment success rate or the notification rate. An inverse correlation is seen between prevalence of resistance and MDR to both the treatment success rate and the notification rate. Source: prevalence of resistance as reported by the WHO/IUATLD Global Project on Anti-tuberculosis drug resistance surveillance, from (WHO/IUATLD 1997; WHO/IUATLD 2000); treatment success and notification rates for these regions are from (WHO 2003).

R^2 values: (A) DR Prevalence and Treatment Success Rate, R^2= 0.191. (B) MDR Prevalence and Treatment Success Rate, R^2= 0.0901 (C) DR Prevalence and Notification Rate, R^2= 0.0372. (D) DR Prevalence and Notification Rate, R^2= 0.0028

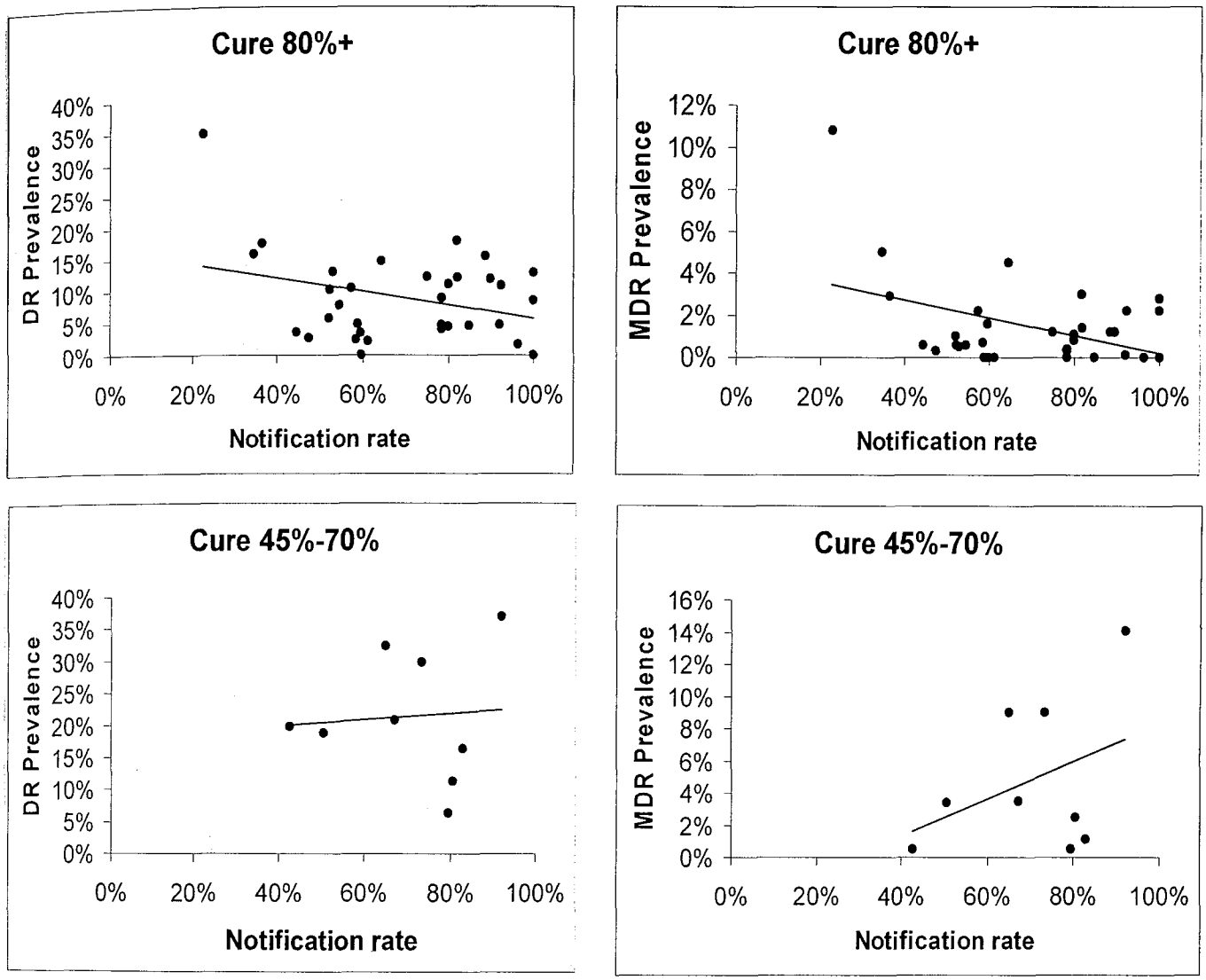


Figure 1-9. ***Resistance prevalence from surveillance by cure and notification rates in program.*** The relationship between resistance prevalence and notification rate is explored. When the cure rates are high (above 80%) in the region, higher notification rates are correlated to a lower prevalence of any resistance (DR) and MDR. In comparison, when the cure rates are low (at a range of 45%-70%) within the region, higher notification rates are correlated to higher prevalence of any resistance (DR) and MDR. *Source:* prevalence of resistance as reported by the WHO/IUATLD Global Project on Anti-tuberculosis drug resistance surveillance, from (WHO/IUATLD 1997; WHO/IUATLD 2000); treatment success and notification rates for these regions are from (WHO 2003)

VIII. Resistant strains transmission

As reviewed in the previous section, recent surveillance programs have documented the emergence of resistance within TB epidemics. To begin to understand the spread of drug resistance, it will be helpful to review efforts to characterize the transmission rates of

drug resistant strains, and how this compares to the spread of other drug sensitive strains within the same population.

The relative transmission rate of resistant, and in particular MDR, strains remains a source of significant uncertainty. Experiments have suggested significant variation in the transmission rates of resistant strains, ranging from reduced to greater than drug sensitive strains. In one study, the contacts of source cases with either sensitive or resistant TB were examined, and similar risks of infection and disease were found amongst contacts of either type. Higher risks of infection were found only amongst resistant cases which were previously treated. (Snider, Kelly et al. 1985) A recent study in Brazil also found similar risks of infection and disease for drug sensitive and resistant source cases, with about 40% of household contacts infected and 4% with active disease. (Teixeira, Perkins et al. 2001) Another study used contact tracing combined with RFLP analysis to show MDR transmitted to contacts at similar rates. In another study, 6% of contacts with MDR patients were found to convert to active disease. (Nitta, Knowles et al. 2002) This can be compared to another study in which 4% of national and 8% of foreign born contacts of drug sensitive cases converted. (Marks, Taylor et al. 2000) These studies together suggest similar rates of conversion amongst contacts of sensitive and resistant source cases.

Studies have also investigated the proportion of resistant cases due to ongoing transmission, by two different methods. The first method involves estimating the prevalence of resistance amongst new and previously treated cases, assuming that new cases are caused by primary resistance, and previously treated strains by resistance acquired during treatment. This classification was used in initial studies of resistance. (WHO/IUATLD 1997) Later studies, using RFLP analysis and contact tracing, revealed that the proportion of acquired resistant cases were overestimated, as some of the previously treated cases appeared to have been initially infected with resistant cases. (Van Rie, Warren et al. 2000) In order to correct for this, later surveillance reports referred to

the proportion of resistant cases amongst new and previously treated cases, without inferring from this the cause of resistance. (WHO/IUATLD 2000) In general, contact tracing or strain typing are required to differentiate acquired and primary resistance.

More recently, convention has led towards using clustering of strains as a surrogate for recent transmission, and such analysis has been used to estimate the relative proportion of resistant strains in a population due to ongoing transmission. In an area of high incidence (Capetown, South Africa), the proportion of resistance due to primary transmission, as detected by RFLP strain typing, was high; over 50% of MDR cases were clustered. (Van Rie, Warren et al. 2000) In Denmark, over 70% of resistant strains were found to be clustered. (Thomsen, Bauer et al. 2000) There have also been reports of 'super fit' resistant strains which have been found more frequently in clusters and are believed to have greater transmission. (Agerton, Valway et al. 1999) (Toungoussova, Sandven et al. 2002) (Kruuner, Hoffner et al. 2001)

Other strain typing studies have found resistant cases less likely to be found in clusters. (Samper, Iglesias et al. 1998; Garcia-Garcia, Jimenez-Corona et al. 2000) (Nitta, Knowles et al. 2002) Cases with a particular INH resistance mutation were found in clusters less frequently than drug sensitive strains. (Van Soolingen, Borgdorff et al. 1999) Further discussion of studies considering the relative fitness of resistant strains, as well as model simulations exploring the implications of varying fitness costs, are found in Appendix 5.

IX. Summary

In this chapter, I reviewed trends in epidemiology of TB, methods for measuring disease burden and surveillance of epidemics, and trends in drug resistance. This sets the scene for the questions posed in this thesis, highlighting the recent (and progressing) state of TB epidemics worldwide. Resistance emerges from treatment patterns, and has been

found worldwide in every region surveyed, with small number of regions showing hotspots, or highly elevated levels of, drug resistance. I also review studies characterizing the transmission rate of resistant strains, and comparison to drug sensitive strains, and discuss why this is of importance to understanding resistant epidemics. In Chapters 5-9, I develop models to understand the emergence of resistance within TB epidemics, comparing the trends in resistance available from initial local and gloabal resistance surveillance. The relative fitness of resistant strains is incorporated into the model developed in Chapter 5, and the sensitivity of model to these parameters assumptions are explored in Chapter 6. The effects of relative fitness are also considered in a within-host model for TB infection developed in Appendix 5. To complement these analyses, a full review of control programs, how they have developed over time, and current practices worldwide, is reviewed in Chapter 3. Together with a review of the molecular biology of *M. tuberculosis* in Chapter 2, this forms the basis for the models developed in later chapters.

2. Chapter 2

Molecular biology of tuberculosis

" ... strangely-visited people
All swol'n and ulcerous, pitiful to the eye,
The mere despair of surgery, he cures,
Hanging a golden stamp about their necks,
Put on with holy prayers; and 'tis spoken,
To the succeeding royalty he leaves
The healing benediction..."

-- Shakespeare, in *Macbeth*

2. CHAPTER 2

MOLECULAR BIOLOGY OF TUBERCULOSIS

I. GOALS

II. ABSTRACT

III. LIFECYCLE OF TUBERCULOSIS DISEASE

III. A. TRANSMISSION
III. B. RISK OF TRANSMISSION
III. C. VIRULENCE
III. D. AT RISK FOR INFECTION
III. E. INFECTION
III. F. FORMATION OF THE TUBERCLE
III. G. LATENCY
III. H. PERIOD OF INFECTIOUSNESS

IV. DIAGNOSIS

V. PROPERTIES OF *M. TUBERCULOSIS*

V. A. MORPHOLOGY OF THE TUBERCLE BACILLUS
V. B. VIRULENCE
V. C. DORMANCY

VI. HOST-PARASITE INTERACTION

VI. A. HOST RESISTANCE TO INFECTION
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VI. C. MACROPHAGE INTERACTION
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VI. E. VACCINE

VII. FITNESS OF *M. TUBERCULOSIS* STRAINS

VII. A. RESISTANT MTB STRAINS
VII. B. COMPENSATORY MUTATIONS IN TUBERCULOSIS TREATMENT

VIII. SUMMARY

I. Goals

I. A. To review the molecular biology of tuberculosis.

I. B. To examine the lifecycle, transmission, and virulence of *Mycobacterium tuberculosis*.

II. Abstract

Tuberculosis is an infectious disease caused by *Mycobacterium tuberculosis*. *M. tuberculosis* is an aerobic, non-motile, acid-fast bacillus, with a slow generation time (both *in vitro* and *in vivo*) of about 24 hours. *M. tuberculosis* is transmitted principally through direct contact with a source case that emits infectious droplet nuclei in their sputum. After entering a susceptible host, droplet nuclei lodge in alveolar regions of the lungs. From early after infection, mycobacteria encounter a host immune response. The balance set in this early challenge by the immune system may determine the outcome of infection. In the majority of cases, infection is successfully contained by the immune response, and the initial foci of disease are healed. However, mycobacteria may remain dormant in the body, and this latent infection reactivate years or decades later in reactivation or post-primary disease. In a small proportion of cases, infection leads directly to progression and primary disease. The ability of a strain to successfully infect a susceptible host and cause progressive primary disease is known as virulence.

III. Lifecycle of tuberculosis disease

III. A. Transmission

Tuberculosis is an airborne transmissible disease caused by *Mycobacterium tuberculosis*. Tuberculosis is transmitted directly from patients with active pulmonary

or laryngeal TB to a susceptible individual by the inhalation of small aerosolised infectious droplets. These droplet nuclei are expelled from an infectious individual when they cough or sneeze. (Johnston RF 1974) These droplet nuclei are 1 – 5 microns in diameter, small enough to dry while airborne, contain several tubercle bacilli, and can remain suspended in the air for several hours. A single cough can produce on the order of 3000 infectious droplet nuclei. The air in a closed room can remain infectious even after an infected patient leaves the room. (Haas 2000; Mandell 2000)

More recently, the mode of transmission through airborne droplet nuclei has been characterized through experiments on sputum from infectious patients. Fennelly's cough box has been used to isolate viable airborne mycobacteria emitted in droplet nuclei from infectious cases. In these experiments, patients are asked to cough through into tubing which leads into Fennelly's cough box. The air in the box is then sampled and the size of the particles estimated. These experiments have found a wide range in the number of colony forming units (CFU) generated per patient sample. As expected, smear positive cases generated more CFUs than smear negative. The size of the particles in these experiments was also estimated, and found that 49% of particles which contained bacilli were in the range of 1.1-2.1 μm. (Fennelly and Nardell 1998)

III. B. Risk of transmission

The probability of transmission when another person inhales droplet nuclei is related to several factors, including the duration of exposure, closeness of contact, infectiousness of the TB patient, and the environment of exposure. Brief contact with an infectious individual carries little risk for infection, and prolonged exposure is usually required. Large respiratory secretions do not factor into the transmission of TB, as they are removed by mucociliary action and coughing.

After inhalation, the droplet nuclei travel to the alveolar region. However, the majority of the droplet nuclei are trapped in the airways, and are removed or destroyed in the respiratory tract. Only the smallest of the droplet nuclei, containing about 1 to 2 tubercle bacilli, reach the alveolar space. Successful human infection is estimated to require from 5 to 200 tubercle bacilli. This number is a function of virulence of the tubercle bacilli and the host microbactericidal activity of alveolar macrophages.

Transmission occurs through prolonged intimate contact with an infectious case. (Grzybowski, Barnett et al. 1975) Most of transmission occurs from sputum positive cases with extensive pulmonary disease and cavitary lesions. (Iseman 2000) The studies first supporting transmission by airborne droplet nuclei, and quantifying the risks of transmission, were performed in the 1970s. (Rouillon, Perdrizet et al. 1976; Iseman 2000) Contact investigation studies have allowed the analysis of the risk of transmission of infection and disease to contacts of cases. Higher rates of both infection and disease have been associated with close contact to a smear positive index case.

Contact investigation studies have shown that the risk of transmission is correlated to the smear status of the index case, with higher rates of transmission from smear positive cases. (Rouillon, Perdrizet et al. 1976; Rose, Zerbe et al. 1979) In one study in England in 1954, 65% of household children contacts of smear positive culture positive cases were infected, with 27% when the index case was smear negative culture positive. (Shaw and Wynn-Williams 1954) In another study in Canada, 45% of household contacts of smear and culture positive cases were infected, while 26% were infected when the index case was smear negative culture positive. (Grzybowski and Allen 1964)

The risk of infection also depends upon the duration and intensity of contact. A common distinction is household and casual contacts; household contacts experience a

much higher risk of infection. In one such study, when the index case was smear positive, 46% of household contacts were infected, while 34% of non household contacts were found infected. When the index case was smear negative but culture positive, 28% of household contacts and 24% of non-household contacts were found infected. (Rose, Zerbe et al. 1979) Another study in the Netherlands gave similar results: when the index case was smear and culture positive, 20.2% of household contacts were infected, while 3.7% of casual contacts were infected. When the index case was smear negative but culture positive, 1.1% of household contacts were infected, while 0.2% of casual contacts were infected. (Loudon and Romans 1969)

Another study in Canada from 1966-1971 examined both the risks of infection and disease amongst household and casual contacts of smear positive or negative cases. This study found higher rates of infection amongst older contacts, amongst household contacts, and amongst contacts of smear positive index cases. The risk of infection due to close contact with an active smear positive case was 27%, compared to 12% from casual contact. However, amongst contacts of the older age groups, there was also less of a difference between the rates of infection amongst contacts of smear positive and negative index cases. This suggests that the higher infection rates in the older age groups are partially attributable to risks of lifelong exposure and reactivation of infection acquired previously from another source. This study also examined the rates of disease amongst contacts, and found higher rates of disease amongst younger contacts, amongst close contacts, and amongst contacts of smear positive index cases. (Grzybowski, Barnett et al. 1975) In a more recent study, contacts of index cases were examined according to grade of sputum smear positivity. 609 contacts of 134 index cases were traced over two years. This revealed four new cases of active disease, each a close contact of a highly smear positive index case. (Liippo, Kulmala et al. 1993)

III. C. Virulence

Virulence describes the capacity of the bacterial strain to infect a host and produce active progressive disease. Infectivity describes the likelihood of the strain to cause an infection given exposure to a susceptible host. Given the difficulty in measuring and quantifying extent of exposure, it has not been possible to separate the strain infectivity from disease progression. Together, the ability of the strain to infect a susceptible host and to cause progressive disease is referred to as virulence. (Iseman 2000) Mycobacterial strain virulence has been tested in animal models. Typical measurements of strain virulence in animal models include number of bacilli in organs, severity of tissue damage, and extent of dissemination.

III. D. At risk for infection

Close contacts of an infectious tuberculosis patient are at highest risk for TB infection. The risk of infection is greater when the contact is emitting a higher number of mycobacteria in their sputum: thus higher risks are found for active patients with positive acid fast smears, as opposed to only positive cultures. Rates of infection are about 27% for close (household) contacts of infectious TB patients. (Haas 2000) (American Thoracic Society 1992)

III. E. Infection

In the alveolus, the tubercle bacilli begin to compete against the host resistance. The bacilli are phagocytosed by non-activated alveolar macrophages. The intracellular bacilli either multiply or are destroyed, depending on the virulence of the bacilli and the innate microbacteriocidal activity of the macrophage. The tubercle bacilli appear to have evolved a mechanism to largely subvert the killing action of the macrophage. Infected macrophages also produce and release chemotactic factors that attract more

macrophages and monocytes, as well as inflammatory cytokines that activate complement. Infected macrophages may also die, releasing bacilli and attracting more monocytes.

Secondary sites of infection begin to form about two weeks after the initial infection. A small number of the infected alveolar macrophages begin to spread through lymphatic channels to regional lymph nodes, and through the bloodstream to other tissues and organs. The areas where TB is most likely to develop are the lungs, kidneys, brain, and bone.

III. F.Formation of the tubercle

Over the course of a month after the initial infection, the immune system responds to the tubercle bacillus by the formation of small nodules called tubercle granulomas. The formation of these tubercle contributes to the halting of the logarithmic multiplication of the bacillus, the decrease in bacillary numbers, and the control of infection. The tubercle is formed by rapid and sustained recruitment of T cells and monocytes. The tubercle has central cavitation and liquefaction; cavity formation occurs with massive tissue necrosis stimulated by cell mediated cytotoxicity. Bacilli are found both extracellularly in liquefied areas and intracellularly in tissue macrophages. The end result of this process of tubercle formation, which is driven by antigen, is the control of bacillary multiplication.

The primary and secondary sites of TB infection are rarely sterilized, even during the lifetime of the host. The persistence of viable bacilli in the sites of infection grants the host immunologic responsiveness as well as immunity to superinfection. Thus, hosts that retain viable bacteria will rapidly eliminate a new infection. If, however, one manages to completely sterilize an infection by use of heavy

chemotherapy, such that no viable bacilli remains in the host, then the hypersensitivity and immunity will be lost within a few months.

III. G. Latency

TB infection can progress to TB disease when the immune system fails to keep in check the growth of the tubercle bacilli. This progression to disease occurs in approximately 10% of individuals infected with the tubercle bacilli. Of this 10%, approximately half of the infected individuals will develop the disease within the first two years after infection (*primary tuberculosis*), and another half later in their lives (*reactivation tuberculosis*).

The risks of progression to active disease are greatest soon after infection, declining exponentially with time since initial infection. About 1.5% of cases progress to active disease within the first year after infection. Within five years after infection, the cumulative risk for progression is 5 to 10%. The remaining lifetime risk for progression is about 5 to 10%. (Enarson and Roullin 1998) (American Thoracic Society 1990)

III. H. Period of infectiousness

A patient with pulmonary tuberculosis is infectious during the period when they are emitting infectious droplet nuclei. Thus, in order to be infectious, the bacterial load within a patient must reach a certain level and must be located within the cavities of the lung where there is access to the airways. A patient is considered to be infectious if (and only if) they test sputum smear-positive. A typical sputum test is sensitive to detect a minimum of 5,000 – 10,000 acid fast bacilli (AFB) / mL sputum. (Levi 1995)

The degree of infectiousness declines rapidly as appropriate treatment therapy is started. Usually within two weeks of initiating chemotherapy, patients will become

non-infectious, as their cough and concentration of bacilli in sputum decreases. Thus, a patient is rendered non-infectious soon after the start of effective treatment. The time from initiation of treatment to culture conversion and presumed non-infectiousness may vary. Early studies suggested that within a month after the start of treatment organisms were non viable. While cases may remain smear positive up to 6 months after the start of treatment, the culture should turn negative after a month. Cases remaining culture positive after 3 months should be suspected of treatment failure and resistance. (Harris and Karakusis 1979) More recent studies have also shown that most cases convert to culture negative status within 4 to 8 weeks (75% of cases). (Cohn, Catlin et al. 1990) In terms of clinical treatment practices, the normal program practices consider that three consecutive negative smear positives are correlated to very low probability of transmission. (American Thoracic Society 1992)

IV. Diagnosis

Tuberculosis is clinically diagnosed by a sputum test. Sputum is taken from the patient and cultured to test for the presence of mycobacteria. The test is not specific to *M. tuberculosis*, and therefore will also detect other mycobacteria. The sputum test can give a false positive for tuberculosis if there is a large presence of other mycobacteria in the environment. The sputum smear test can detect only active cases of tuberculosis. When an infected patient becomes infectious (the mycobacterial load exceeds a threshold, and has access to the airways) mycobacteria will be emitted in the sputum and detectable in culture. The limitations of this common diagnostic are clear: infected patients cannot be diagnosed until they become infectious; lack of specificity gives false positives; there is no quantitative measure for the bacterial load of the infection. (Allen BW 1992; Levi 1995)

A TB skin test is used to test for infection with *M. tuberculosis*. This skin test detects reactivity to tuberculin, a mixture of mycobacterial antigens. (Bothamley 1991) To diagnose infection with tuberculosis, tuberculin is injected into the skin and the induration response is measured at 2 or 3 days. The test is read by observing the size of induration at the site of contact with the tuberculin. The extent of induration reflects the patient's immune reaction to the mycobacteria. (Bothamley 1999) A test is classified according to the patient's risk group, such that a larger induration is required of those individuals who have fewer known risk factors for tuberculosis. An induration of > 5 mm is considered positive for individuals in contact with active cases of tuberculosis, or who are infected with HIV. An induration of > 10 mm is considered positive for those of middle-risk for tuberculosis, such as residents of an area with a high prevalence. An induration of > 15mm is required for individuals with no known risk factors for tuberculosis. (Bothamley 1999)

The induration response to the application of tuberculin represents the development of delayed-type hypersensitivity (DTH). Macrophages and T cells that have been previously exposed to mycobacterial antigens will rapidly respond to the presence of these antigens on the skin. Memory T cells, responsive to *M. tuberculosis*, will gather at the site of the skin infection and will activate the release of cytokines and proliferation of immune cells. False-positive skin tests can be produced by exposure to other cross-reacting mycobacteria present in the environment. Infected patients may also test negative (a false-negative) for the skin test if complications, such as progression of AIDS, interfere with the TH1 immune response. (Bothamley 1991)

V. Properties of *M. tuberculosis*

V. A. Morphology of the tubercle bacillus

The *M. tuberculosis* bacillus is a member of the genus Mycobacterium. There are some 55 species of mycobacterium, but those most closely phyologenetically related to *M. tuberculosis* are known as the *M. tuberculosis* complex and include *M. bovis*, *M. microti*, and *M. africanum*. (Fishman 1988; Gangardharam and Jenkins 1997; Iseman 2000)

The most distinguishing characteristic of the microbes in the *M. tuberculosis* complex is their acid-fast property. These mycobacteria resist staining with traditional stains, including the Gram stain, but retain dyes which have penetrated the cell wall. This is due to the high lipid content of the mycobacterial cell wall. The mycobacterial cell wall is lipid rich, containing waxes and glycolipids that account for about 60% of the cell as dry weight. This cell wall gives mycobacteria a waxy coat that serves both as a defence and to retain fluid. This may contribute to ability of mcyobacteria to remain viable for long periods. The primary components of the cell wall include peptidoglycans, arabinogalactans, and mycolic acid. The long chain mycolic acids, which are unique to mycobacterial cell walls, contribute to the acid fastness of the cell. (Levi 1995; Davies 1998; Iseman 2000)

M. tuberculosis is slow growing, with a generation time *in vitro* of about 24 hours. This generation time depends on environment and culture technique. Visible colony growth on solid culture medium takes at least three to six weeks to appear. Cultured *in vitro*, *M. tuberculosis* appears rod shaped and slightly curved or straight. The mycobacterium also appears smaller than most bacterial pathogens, about 1 to 4 mm in length and 0.3 to 0.6 mm in diameter. The mycobacteria are also observed to grow in dense clusters referred to as 'cords' because the bacteria are aligned in parallel. *M. tuberculosis* is an aerobic bacillus, whose growth and virulence appears to be related to the availability and use of oxygen. When deprived of oxygen, mycobacteria have

been shown to remain viable, but have slowed or ceased replication. (Levi 1995; Davies 1998; Iseman 2000; Mandell 2000)

V. B. Virulence

Bacterial virulence refers to the ability of a pathogenic organism to cause disease. For mycobacteria, virulence depends upon factors which influence the ability to infect a human host, to enter and survive within macrophages, and to overcome immune defences and cause active disease. (Grange 1998)

The virulence of a tuberculosis strain depends on its ability to survive host defences. Virulence of *M. tuberculosis* is thought to be multifactorial, with a complex interaction between component factors determining virulence. (Gordon 1996) A key determinant of mycobacterial virulence is the ability to enter host macrophages and persist in the typically acidic environment. Mechanisms of this persistence have been shown to include an avoidance of the endocytic pathway and localization within the macrophage to non-acidic vacuoles. (Barker, George et al. 1997) The ability to replicate within macrophages has also been shown necessary for mycobacterial virulence. Studies have examined the genes selectively expressed in mycobacteria growing within macrophages. The genes found to be upregulated in mycobateria found within macrophages include those functioning in protection from oxidative stress and nutrient utilization. (Triccas 1999) These findings suggest that mycobacteria upregulates genes to enhance its survival within the macrophage environment.

V. C. Dormancy

M. tuberculosis has the ability to remain dormant for extended periods of time. (Koch 1997) The biological nature of this dormancy is not well understood. However, this feature is central to allowing bacteria to remain in a host for years following the

initial infection, a state referred to as latent tuberculosis. (Colston 1999) This makes bacterial sterilization of a host very difficult, particularly since many of the antituberculosis drugs are active only on dividing cells. (Smith 1994) Drug treatment will usually kill most of the mycobacterial population within several weeks; however, mycobacterial persisters can evade the drug by entering into a dormant state and remain in the host. These remaining bacilli can later lead to reactivation of the infection and post-primary tuberculosis. (Colston 1999)

VI. Host-Parasite Interaction

Infection begins as a susceptible host inhales droplet nuclei containing mycobacterium. Successful droplet nuclei will lodge in alveoli and air passages of the lung, establishing the initial site of infection. This is referred to as the Ghon focus, or the local focus of disease. From this initial focus, mycobacteria are transported to lymph nodes in the long, where foci of disease develop. Together, the Ghon focus and the foci established in the lung are referred to as the primary complex. From this initial primary complex of disease, mycobacteria may continue to disseminate further in the body, when carried by the lymphatic and blood streams to other organs. In most cases, the primary infection is resolved by host defences, and individuals remain latently infected but do not directly progress to active disease. The early host immune response may resolve the initial infection but leave mycobacteria in the primary complex dormant; in about 10% of infections, cases reactivated to post-primary disease decades after the initial infection. (Dannenberg 1998; Davies 1998)

The time table for development of tuberculosis disease was described by Koch as occurring in 4 stages. In the first stage of disease, lasting from 3 to 8 weeks after initial infection, and includes the development of a primary complex of disease. During this stage, there is phagocytosis into macrophages, and most of the

mycobacteria from the initial infection likely die. The remaining mycobacteria are able to proliferate within the macrophages. In this stage the early cellular immune response also begins, and there is a conversion to tuberculin positivity. In the second stage, lasting up to 2 months, there is continued dissemination of the bacteria from the primary focus, and the disease may spread to other locations and organs. During this stage, the remaining mycobacteria replicate within macrophages, and cause cell death. The immune response also continues: chemotactic factors and complement are activated, and monocytes are recruited. In the third stage, lasting 3 to 4 months, there is continued spread of disease from the primary focus. In the fourth stage, lasting up to 3 years, there is resolution of the primary complex, and continued development of more slowly growing extrapulmonary lesions. (Dannenberg 1998; Davies 1998)

Reactivation, or secondary tuberculosis infection, usually begins in the apical or posterior segments of the upper lobes. The infection is believed to originate in these locations because of the favourable environment for tubercle bacilli multiplication. As an obligate aerobe, the tubercle bacillus reproduces best in a high oxygen environment. The relatively high partial pressure of oxygen in the upper portions of the lungs is particularly suitable for the tubercle bacilli lifestyle. (Dannenberg 1998; Davies 1998; Grange 1998)

VI. A. Host Resistance to Infection

Numerous studies have suggested that individuals vary in susceptibility to tuberculosis disease, and that genetic variation is important in determining the outcome of infection. (Skamene 1991) (Hoal 2002; Malik and Schurr 2002) Whole-genome linkage analyses in families with multiple cases have also been useful in identifying factors associated with susceptibility. (Hoal 2002) Sibpair studies have found that host genetic factors can be important determinants of susceptibility to infectious diseases,

including tuberculosis. Mutations in interferon gamma receptor lead to increased susceptibility to mycobacteria and BCG infection in homozygous children. Variation in the vitamin D receptor has also been associated with susceptibility. (Hill 1998) In one study, families with two siblings affected by tuberculosis were examined with a genome-wide linkage study to identify genes that affect susceptibility. Two loci showed repeated coinheritance with susceptibility. (Bellamy, Beyers et al. 2000) In other family studies, mutations in genes for interferon-gamma receptor and the interleukin-12 receptor have been correlated to increased susceptibility. This suggests that IFN-gamma and IL-12 may be involved in the pathways for resistance to infection. In particular, cytokines in pathways for macrophage upregulation have been implicated. The absence of key cytokines in children has been associated with susceptibility to lethal mycobacterial infections. It is suggested that mutations in these cytokines may have more subtle affects on these pathways that could influence susceptibility in the adult population. (Levin and Newport 1999)

Use of animal models of tuberculosis, in particular the mouse, has provided valuable contributions in identifying candidate genes involved in resistance to infection. Studies using mouse models of tuberculosis have found that susceptibility to infection and disease is a complex genetic trait. (Schurr, Malo et al. 1991) (Kramnik, Demant et al. 1998) Several genes have been correlated to susceptibility to disease in mice, including the Nramp1 (natural-resistance-associated macrophage protein 1) gene and sst1 (susceptibility to tuberculosis 1) gene. Variation at the sst1 locus has been correlated to growth rates of virulent mycobacteria in the lungs. (Kramnik, Dietrich et al. 2000)

Nramp1 was first isolated by positional cloning of a locus found associated with host resistance. The Bcg gene was known to segregate as a single dominant autosomal gene and to control susceptibility to mycobacteria. Expression of the Bcg gene was

known to activate macrophages to destroy ingested intracellular parasites as part of the early nonspecific immune response shortly after infection. The Bcg gene was mapped using positional cloning in mice, and overlapped a region that included the Nramp gene. (Skamene 1994) Mutations in Nramp1 were sufficient to reduce mice resistance to infections with mycobacterium, as well as salmonella and leishmania. (Canonne-Hergaux, Gruenheid et al. 1999) An Nramp1 homologue, known as NRAMP1, has been identified in humans and found associated with susceptibility to tuberculosis. (Liu, Fujiwara et al. 1995; Bellamy, Ruwende et al. 1998) Nramp1 has been characterized as an integral membrane protein expressed in the lysosomal compartment of macrophages. Nramp1 is thought to be involved with regulating mycobacterial replication after phagocytosis into macrophages. (Canonne-Hergaux, Gruenheid et al. 1999; Buu, Sanchez et al. 2000)

VI. B. Immunology

The tubercle bacteria first encounters host immune system in the form of alveolar macrophages, which phagocytose and carry the bacteria to lymph nodes. For the first several weeks after the initial infection, the bacteria grow exponentially. After several weeks, two components of the cell-mediated immunity develop: delayed type hypersensitivity (DTH) and protective cell mediated immunity (CMI). These two components are thought to be closely associated, and seem to occur at the same time after infection, coinciding with conversion to a positive tuberculin skin test. The development of protective immunity is a result of TH1 activation, while a TH2 or mixed TH1/TH2 response leads to DTH and progression of disease. The development of skin hypersensitivity occurs with the establishment of memory T cells that recognize the mycobacterial antigens. (Grange 1998)

VI. C. Macrophage Interaction

The ability to survive in the macrophage is essential to the ability of *M. tuberculosis* to successfully infect a host. A strain that is unable to survive within the macrophage environment will be cleared by the host immune system. Upon encountering the macrophage, the bacilli is phagocytosed inside the cell and carried within a compartment known as a phagosome. The components of this compartment are destined for degradation, which is accomplished by fusion of the lysosome with the phagosome. The lysosomal enzymes are released into the phagosome, and degrade its components. The lysosome is maintained at a higher pH, required for the lysosomal enzymes to function. (Grange 1998)

In order to survive, the bacilli must evade the macrophage defences. There are two primary mechanisms that *M. tuberculosis* uses to evade these defences. The first is by preventing the fusion of the phagosome to the lysosome. The second is preventing the acidification of the phagosome, so that the lysosomal enzymes will remain inactive. The cellular location of the bacilli within the macrophage determines the nature of the immune response. Bacterial antigens contained within the macrophage phagosomes are presented by MHC-II to CD4+ T cells, which activate the proliferation of the macrophage. In contrast, bacterial antigens present within the cytoplasm of the macrophage are presented by MHC-I to CD8+ T cells, which in turn lyse the macrophage. (Grange 1998)

VI. D. TH1 and TII2 responses

One of the key immunologic features of the pathogenesis of tuberculosis is the interplay between two classes of CD4+ T helper cells, TH1 and TH2. Depending upon which prevails in a given infection, the mycobacterial population will either be cleared, form a latent infection, or form primary tuberculosis. Studies have examined the

factors that influence the dominance of either TH1 or TH2. Correlations between development of active disease and the balance between TH1 and TH2 response have been shown. Cases with active disease have excess expression of cytokines in the TH1 pathway. In comparison, individuals with latent infection show higher expression of cytokines in the TH2 pathway.

Cytokines of either group were found to cross-regulate and inhibit the proliferation and activation of those in the other group. TH1 cells activated against *M. tuberculosis* produce the cytokines IL2 and IFNγ, promoting protective immunity. Activated TH2 cells produce IL4, IL5, IL6, and IL10 cytokines, promoting tissue destruction. When the TH1 response dominates the response to an infection, granuloma formation and protective immunity result. When either a TH2 or a mixed TH1/TH2 response exists, the immune response will lead to the development of hypersensitivity and the progression of disease. (Grange 1998) The cytokines produced by either pathway inhibit the maturation of cells from the other pathway, thus locking in a given TH1 or TH2 response. For example, the TH2 cytokine IL10 inhibits the TH1 maturation pathway. (Mossman 1991)

VI. E. Vaccine

The common vaccine against tuberculosis is BCG, which was first derived from an avirulent strain of bovine *M. tuberculosis*. BCG provides protective immunity by activating macrophages to recognize the tubercle bacilli. When an immune individual is infected with tuberculosis, they will rapidly mount an immune response to clear the infection.

However, the efficacy of BCG vaccination is found to vary greatly (from 80% to 0%) in different trials. (Fine 1995) This variation has been best explained by regional differences in environmental mycobacteria. Depending on the prevalence and

type of local mycobacteria in the environment, a population may be at higher risk for sensitization to mycobacteria and development of either a TH1 or a TH2 response. The BCG vaccine acts by promoting the existing T helper cell pattern, such that it will imprint either a TH1 (protective) or TH2 (tissue-damaging susceptibility) response. (Bretscher 1991) However, in those individuals not previously sensitized to mycobacteria, BCG vaccination will produce a TH1 response, affording protection to tuberculosis infection. (Ten Dam HG 1980) Thus, BCG is often given to the young, particularly in regions where the efficacy of vaccination is likely to decrease with age.

VII. Fitness of *M. tuberculosis* strains

The resistance to treatment with antibacterial drugs has often been found associated with a fitness cost to the resistant organism. The fitness of an organism has several important components, including: the growth rate (births and deaths) both within an infected host and within the environment, the transmission rate for infection of new hosts, and the clearance rate from an infected host. The fitness cost of resistance is measured by comparing these rates for sensitive and resistant strains.

The experimental evidence to support a fitness cost of resistance comes from a number of organisms and suggests that this cost is widespread. Such experiments have measured the relative growth rates of sensitive and resistant strains *in vitro* (through competition experiments). (Andersson and Levin 1999) Other experiments have looked retrospectively at previously collected data from patients and examined the correlation between sensitive and resistant frequencies to the rate of antibiotic consumption. (Austin, Kakehashi et al. 1997)

Fitness costs of plasmid-encoded resistance have been studied in *E. coli*. *E. coli* carrying plasmids with resistance genes have been found to have decreased growth rate compared to organisms free of this plasmid. Bacteria grown carrying a resistance

plasmid have been found to acquire chromosomal mutations that restored the growth rate. (Lenski, Simpson et al. 1994; Lenski 1998; Lenski, Mongold et al. 1998)

A fitness cost has also been found associated with mutants resistant to streptomycin. The drug acts by interfering with protein synthesis, targeting the bacterial ribosome. (Goodman 1996) The most common mechanism of streptomycin resistance is the mutation of genes affecting the bacterial ribosome, which prevent the enzymatic inhibitory action of streptomycin. The primary mutation found in human tuberculosis cases resistant to streptomycin is a point mutation that alters the ribosomal binding site. (Goodman 1996) The ribosomal mutations (in the *rpsL* gene) causing streptomycin resistance reduce the efficiency of translation. The reduced translation rate is thought to result in the reduced fitness of the streptomycin resistant bacteria compared to wild type. (Bohman, Ruusala et al. 1984)

However, there also is some evidence suggesting the existence of 'no cost' resistance mutations. For example, resistance to high concentrations of streptomycin, conferred by a substitution at the 42nd codon of the *rpsL* gene in *S. typhimurium*, shows fitness similar (or even slightly higher than) wild type. Other mutations found to cause resistance to streptomycin have been shown to confer fitness disadvantages. Thus, the fitness cost of drug resistance appears to be allele-specific. (Andersson and Levin 1999)

VII. A. Resistant MTb strains

In *M. tuberculosis* strains, there is some, although limited, experimental evidence to suggest a fitness cost associated with developing antibacterial resistance. Tubercle bacilli resistant to isoniazid have shown reduced fitness *in vitro*. Whether this holds true *in vivo* is unclear. There is limited experimental evidence to suggest how resistance to other common tuberculosis antibiotics affects strain fitness.

There is also evidence that resistance to isoniazid may be correlated with decreased virulence. Although INH resistance does not seem to affect bacterial replication in monocytes *in vitro*, these mutations do confer increased sensitivity to reactive oxygen. As tubercle bacteria are exposed to high levels of oxygen during the course of an infection, the resistant mutants may be more compromised by the environmental conditions present *in vivo*. Resistance to isoniazid may be deleterious to the response of the pathogen to the immunological defence of the host cells. (Davies 1998)

There also appears to be an important role for compensatory mutations in the fitness of resistant strains, both *in vitro* and *in vivo*. *M. tuberculosis* strains resistant to isoniazid, which normally show reduced fitness *in vitro*, have been found to have increased fitness when associated with a second-site mutation in the *katG* gene. This intragenic suppressor mutation appears to serve as a compensatory mutation.

A study has also examined deletions of the *katG* gene, which confers high resistance to isoniazid, and is associated with decreased strain fitness. The loss of the *katG* gene is believed to cause the loss of catalase activity as well as bacterial virulence. Such *ΔkatG* mutants were found to rapidly acquire second-site compensatory mutations that restored virulence while maintaining the high level of isoniazid resistance. The compensatory mutation was mapped to the promoter region of the *aphC* gene. The mutation increased transcription from this promoter, resulting in increased levels of the AphCp. The over-expression of this alkyl hydroxy peroxidase is believed to compensate for the effects of the decreased catalase activity. (Sherman, Mdluli et al. 1996; Heym, Stavropoulos et al. 1997) However, more recent experimental evidence has suggested that the over expression of *aphC* (on a plasmid) will not restore the virulence of a *katG* mutant. The overexpression of *aphC* was not seen to affect the level of resistance of bacteria, in either a *katG* wild type or mutant background. (Heym,

Stavropoulos et al. 1997) The mechanism for the compensatory mutation in the *aphC* gene remains unclear; it is possible that the mutation must be chromosomal (as opposed to carried on a plasmid) to restore virulence. It is also possible that there is an additional factor that is regulated by this mutation, which is responsible for the restored virulence.

VII. B. Compensatory mutations in tuberculosis treatment

The fitness of the resistant strain, and the evolution of compensatory mutations that may increase competitive fitness, are important considerations in the design of treatment protocols. A common suggestion to the public-health problem of epidemics of antibiotic resistance is to reduce antibiotic consumption. Such an argument presupposes that in the absence of selective pressure of the antibiotic, the resistant organism will be less fit than the corresponding sensitive strain. Indeed, if this were always the case, such a solution would be tenable. When the sensitive strain maintains a fitness advantage in the absence of drug selection, under the appropriate conditions, removing the selection will allow for the re-establishment of a sensitive population. The timescale for the reversion of the population to sensitivity should be similar to the time taken for the emergence of resistance.

However, when the resistant strain adapts to its environment through the acquisition of compensatory mutations that increase its fitness, this no longer holds true. Once the drug selection is removed, the compensated resistant strain retains its fitness advantage. And if the fitness of the resistant is greater than that of the sensitive, then simply removing the drug selection is unlikely to allow for reversion to a sensitive population. Even if the fitness of the resistant strain with the compensatory mutation is not greater than that of the sensitive strain, in the absence of drug, the increased fitness

of the resistant will mean that the timescale for the replacement of the resistant population with sensitive organisms will be longer than the converse.

VIII. Summary

In this chapter, I review the molecular biology of *M. tuberculosis*. The key elements of MTb reviewed here include lifecycle, transmission and virulence. Also briefly discussed is the diagnosis of infection, but more details on diagnostic tests for active cases can be found in Chapter 3, which reviews TB treatment. Understanding these properties of MTb sets the stage for exploring the nature of TB epidemics, and of interest in this work, how to best model the transmission of TB amongst individuals within a population and the impact of chemotherapy and other elements of a control program. This also provides introduction to the basis for the models developed in Chapter 5 for transmission and treatment, as well as within-host models considered in Appendix 5.

3. Chapter 3

Treatment of tuberculosis

"It happens then as it does to physicians in the treatment of Consumption, which in the commencement is easy to cure and difficult to understand; but when it has neither been discovered in due time nor treated upon a proper principle, it becomes easy to understand and difficult to cure. The same thing happens in state affairs; by foreseeing them at a distance, which is only done by men of talents, the evils which might arise from them are soon cured; but when, from want of foresight, they are suffered to increase to such a height that they are perceptible to everyone, there is no longer any remedy."

-- Machiavelli

3. CHAPTER 3

TREATMENT OF TUBERCULOSIS

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XI. SUMMARY

I. Goals

I. A. To review the literature on the treatment of tuberculosis.

II. Abstract

The introduction of effective chemotherapy for tuberculosis in the 1960s allowed for the rapid cure of patients. The first effect of introducing TB programs which included chemotherapy was the rapid reduction in mortality, while allowing infectious cases to live longer. The early regimens often included only one or two drugs, allowing for rapid selection for drug resistance. The introduction of rifampicin in the late 1960s and the use of multi-drug regimens reduced the rate of emergence of resistance. The establishment of short course chemotherapy (SCC) and national treatment programs under the WHO guidelines for DOTS (Directly observed treatment, short course chemotherapy) standardized treatment practices. The DOTS guidelines proscribe treatment of a new case with at least four drugs for 6 to 9 months. DOTS programs have been shown to rapidly reduce the incidence and death rates.

The detection of infectious cases is central to any treatment program, and accordingly is a cornerstone to improvements in TB control programs. Traditional methods for case detection include the smear and culture tests. Newer methods include the more rapid and automated radiometric culture systems such as BACTEC, the non-radiometric system MB/BacT, and various serological or molecular methods including PCR-based analysis and DNA chips.

The continued emergence of resistance under these programs, has led to a growing need for drug susceptibility testing (DST). Traditional methods for DST include culturing sputum samples and determining resistance by the absolute concentration, ratio, or proportion methods. Newer methods for DST include

automated radiometric systems such as BACTEC, which allow for faster culture with higher sensitivity. In addition, the development of molecular methods using PCR based systems has provided a fast and sensitive alternative test. These tests require knowledge of the genetic basis of resistance and rely on sequencing or hybridization to search for particular mutations associated with resistance.

III. Progress in tuberculosis treatment: a historical perspective

Prior to the use of chemotherapy to treat tuberculosis, cases were treated largely by in sanatoria. Confinement in sanatoria served to reduce the spread and transmission of disease, as well as to offer patients the best opportunity available for cure. This included bed rest, a nutritious diet and fresh air. In addition, some surgical methods and collapse therapy were used for treatment. Treatment of tuberculosis with chemotherapeutic regimens began with the discovery of streptomycin in the 1940s. Treatment with either streptomycin or PAS resulted in a more rapid cure than achieved in sanatorium. Chemotherapy led to sputum conversion as well as clinical improvement after 2 to 3 months of treatment with either streptomycin or PAS. However, the effects of treatment did not appear to be long-lived. There were high rates of relapse to disease shortly after treatment was stopped. (Medical Research Council 1948) Use of combined chemotherapy with both streptomycin and PAS prevented the emergence resistance to either drug, and the cure rates were better than for use of either drug alone. (Medical Research Council 1950)

Isoniazid was introduced for use in the treatment of tuberculosis in 1952. Treatment regimens used in clinical trials by the MRC in 1955 included either isoniazid alone, or in combination with streptomycin or PAS. These trials also confirmed that treatment with a single drug resulted in high rates of drug resistance. (Medical

Research Council 1955) Standard regimens for treatment were developed over the following decade. The standard regimen included streptomycin, isoniazid, and PAS given for from 18 months to 2 years. There were difficulties with these early regimens, including high rates of adverse reactions, patient default, poor adherence, failure, and resistance. (Crofton 1960; BMRC 1973) In the early 1960s, studies showed that home treatment was effective. With the successes of these chemotherapy regimens, treatment in sanatorium became less important. (Tuberculosis Chemotherapy Centre 1959) The introduction of rifampicin in the 1970s provided great improvements to treatment. Including rifampicin allowed regimens to be shortened to 6 to 9 months, referred to as short course chemotherapy (SCC). (Fox and Mitchison 1975; Fox and Mitchison 1976)

The emergence of cases resistant to tuberculosis drugs was seen shortly after the chemotherapy was first used in treatment. When regimens of monotherapy were common in treatment, resistance was seen to rapidly emerge. The introduction of treatment into a population often led to an increase in case duration of infectiousness, creating chronic infectious cases which were unresponsive to treatment. Many of these cases turned out have drug resistance. With the introduction of rifampicin to traditional treatment, there were significant improvements in treatment cure and a reduction in the amount of chronic cases produced. (Lutwick 1995)

IV. Treatment standards

IV. A. Aims

The aims of treatment regimens are to reduce case mortality and infectiousness, stop cycles of transmission, and decrease incidence and burden of disease.

IV. B. Treatment of smear positive cases

A number of different effective regimens for treatment of smear positive pulmonary tuberculosis are found in clinical trials in both developed and developing countries. The typical regimen is given for at least 6 months, the shortest required duration, with a continuation phase of 2 or 3 months. However, longer duration of treatment is required if no pyrazinamide is used in the initial phase or no rifampicin in the continuation phase. Regimens vary by daily or intermittent dosing, self-administered or DOT, regimen and supervision can vary from initial to continuation phase. The relapse rates on these regimens are generally low, below 5%. (Davies 1998; Iseman 2000) These regimens all consist of an initial intensive phase of 2 months and a continuation phase of 4 to 6 months. Fewer drugs are needed for the continuation phase; however, it is critical to include those drugs, including rifampicin, which have strong sterilizing activity. The continuation phase is necessary in order remove remaining bacteria and prevent relapse.

The regimen recommended for use within DOTS programs for treatment of smear positive cases depends upon treatment history. New cases are defined as those which have not received any prior tuberculosis treatment; initial cases are defined as those in which prior treatment may be suspected but not confirmed. (WHO 1999; WHO 2003) The recommended regimen for new smear positive cases includes an initial phase with at least four of the first line drugs, including isoniazid, rifampicin, pyrazinamide, ethambutol, and streptomycin. Proper administration of this regimen to a drug sensitive smear positive patient results in a rapid reduction in bacterial load, reduction in symptoms, and conversion to smear negative within several weeks. During the continuation phase at least two drugs, including rifampicin, are given. (WHO 2003)

Retreatment cases are defined as those that have received at least one month of previous antituberculosis treatment, which remain smear or culture positive. This class includes cases which have failed treatment, relapsed, or defaulted. The recommended standardized retreatment regimen includes five drugs in the initial phase, followed by three drugs in the continuation phase. This regimen can offer high cure rates for drug sensitive cases as well as those with single resistance to isoniazid or streptomycin. However, cases which have developed MDR are most at risk of failure on these retreatment regimens. (WHO 1997; WHO 2003)

IV. C. Treatment of smear negative cases

Many TB programs have questioned whether to treat cases diagnosed with active disease without bacteriological confirmation. A high proportion of cases which are either smear negative or have no mycobacteriological confirmation of disease do receive SCC treatment. (Sbarbaro 1989) When smear samples are unavailable or the smear test inconclusive, treatment may continue as if the test were positive until further results are available. Such presumptive treatment provides caution in treating cases which may be infectious, reducing transmission. However, the treatment of smear negative cases is a secondary priority for a TB program in a high burden country. The first priority is the detection and effective treatment of smear positive cases. Most of transmission occurs from smear positive cases, while the relative contribution of smear negative cases to transmission remains unclear. Further, smear negative cases have fewer symptoms and are harder to diagnose. However, when resources allow, treatment of smear negative cases is important both for curing the patient and for stopping transmission if the case is infectious. (Davies 1998; Colebunders and Bastian 2000; Iseman 2000)

Smear negative cases can be treated effectively with a four month regimen. (Dutt, Moers et al. 1989) Several clinical trials have shown the effectiveness of regimens for treatment of smear negative cases. In one Hong Kong study, 1019 smear negative pulmonary TB cases were followed. 36% of these cases had one or more initial sputum sample culture positive. This study showed that 2-3 months treatment with the basic four drug regimen (isoniazid, rifampicin, streptomycin, pyrazinamide) was not adequate. Relapse rates at 60 months were high; for cases with one culture positive specimen there was a 32% relapse rate, compared to 11% for those culture negative. (Hong Kong Chest Service/Tuberculosis Research Centre/Madras/British Medical Research Council 1984; Hong Kong Chest Service/Tuberculosis Research Centre/Madras/British Medical Research Council 1989) In the second Hong Kong study for treatment of smear negative cases, of 1710 smear negative pulmonary cases, 35% had one or more initial sputum culture positive samples. The cases were given either 3 or 4 month regimens. Amongst cases receiving 3 months regimens of HRZS either daily or three times a week, there was a 7% relapse rate. There was a 4% relapse rate for the same regimen given for 4 months. Amongst cases receiving the 4 month regimen, there was a 2% relapse rate for drug sensitive cases and 8% for cases resistant to H/S but susceptible to R. (Hong Kong Chest Service/Tuberculosis Research Centre/Madras/British Medical Research Council 1989)

In areas with a high prevalence of HIV, coinfection may complicate the detection and treatment of smear negative cases. In such circumstances, it may be advisable to treat HIV coinfected cases with a full course of SCC even when smear negative. (Colebunders and Bastian 2000)

V. Effect of treatment on disease

Effective treatment rapidly reduces the infectiousness of patients. Within two weeks, treatment reduces the frequency of coughing in active cases, reducing the production of droplet nuclei in sputum. (Loudon and Spohn 1969) Under modern treatment regimens, effective treatment is seen to result in smear conversion within several weeks. (Lutwick 1995) The first effect of introducing treatment is to reduce the case mortality. As new programs are introduced, the death rate is reduced, allowing cases to live longer. When this treatment does not result in high cure rates, this may allow for an increase in case duration of infectiousness.

V. A. Deaths under treatment program

Several studies have examined the mortality rates under TB treatment programs in developed countries. In one study in England in 1978-9, amongst 1312 adults with pulmonary TB receiving treatment, 163 (12%) died before completing treatment. Factors correlated to death included smear positive status, extent of cavitation, and age. (Humphries, Byfield et al. 1984) An analysis of another UK study from 1983-1985 found a case fatality rate of 12.9% for adult smear positive cases under treatment. This was ten fold greater than the death rate for age-matched controls. Again, age and extent of disease was correlated to death during treatment. (Cullinan and Meredith 1991)

In program conditions, introducing DOTS SCC has been shown to rapidly reduce the number of deaths due to TB. The implementation of DOTS SCC in high burden countries has reduced the incidence of disease and has prevented a high proportion of deaths. In China, dramatic improvements to control over the past decade, including the implementation of standardized DOTS programs and increased case detection, has significantly reduced incidence. Over 50% of deaths were prevented in this region from 1991-1997, according to a study which estimated the predicted number

of cases and deaths that would have occurred, if such improvements were not implemented. (Dye, Fengzeng et al. 2000) Similarly, in Peru, the introduction and improvements to DOTS programs significantly reduced the incidence of disease. These improvements to control, including increased case detection and standardized treatment with higher cure rates also resulted in reduced mortality. It was estimated that from 1991-2000 nearly 70% of deaths were prevented by implementing these improvements. (Suarez, Watt et al. 2001)

VI. Case Detection

Cases are detected by either passive or active means. Passive case detection refers to symptomatic individuals who report to health authorities seeking treatment. Active case detection entails screening programs to detect either latent infection or active disease amongst a wider population. Such screening may be implemented within select at-risk populations, such as new immigrants, high prevalence communities, and contacts of current cases.

VII. Global Case Detection and DOTS

The majority of global case detection currently occurs through passive means. An estimated 40% of new smear positive cases are detected each year, with about 27 % (1.02 million cases) of these occurring under DOTS programs. DOTS programs have been implemented in 148 countries, and 55% of the world's population lives within regions providing DOTS. The average treatment success under DOTS has been estimated at 80%. (WHO 2002; WHO 2003)

The WHO has set a global target for 2005 of 70% case detection and 85% treatment success. The rate of increase in case finding has stayed similar since 1994; in order to reach detection goals this needs to be increased. The expansion of DOTS

programs has been commonly constrained by factors including resources for staff, laboratories and technical equipment, unregulated private sector, decentralization and poor access to health services. In addition, the expansion of DOTS coverage usually involves notification of cases which would otherwise have been detected under non-DOTS programs. This has meant that the 60% of global cases remaining undetected has not been reduced. National programs have not notified more than 40% of smear positive cases, raising questions about whether some limit of current detection programs has been reached. (WHO 2002; WHO 2003)

VII. A. Active Case Detection

In regions of low incidence, a high proportion of new cases are found amongst high risk groups, including contacts of smear positive cases. Screening contacts of known cases for infection and disease can be an important method for active case detection. Such screening can uncover an emerging epidemic and allow for early prevention. The recommended methods for screening are by concentric circles, such that those with the greatest contact with the source case are screened first. (Narain, Nair et al. 1966) (Grzybowski, Barnett et al. 1975) (Egsmose, Ang'awa et al. 1965; van Geuns, Meijer et al. 1975) (Veen 1992) (Sebek 2000) Close contacts include individuals sharing a household, but may also be found amongst individuals sharing equivalent amounts of time in close proximity in work settings, hospitals, homeless shelters, or even social settings as nightclubs. Casual contacts include most occupational and social contacts. (Chapman and Dyerly 1964) (Grzybowski, Barnett et al. 1975) (Narain, Nair et al. 1966) (Chin, DeRiemer et al. 1998) (Barnes, el-Hajj et al. 1996) (Reichler, Taylor et al. 2002)

While the basis of global TB control is passive case finding, there are also select examples of programs that incorporate active case finding. For example, contact

tracing is commonly employed in the US and UK as a means of identifying infected individuals and new cases. These have been shown to be useful in increasing levels of detection, often reaching cases which otherwise would not have been identified or had access to medical resources. (Menzies 1997; CDC 1998; Murray and Salomon 1998; Broekmans, Migliori et al. 2002)

Recent studies have suggested that contact tracing is an effective means to identify new cases. Retrospective studies have shown that about 10% of new cases were detected through contact tracing, and about 1% of close contacts of a smear positive source case developed active disease. The screening of casual contacts, such as occupational or social contacts spending only short periods in shared spaces, is suggested to be of only limited importance in finding new cases. (Marks, Taylor et al. 2000) A study in San Francisco found that 5% of cases were identified through contact tracing. Of contacts of smear positive cases examined, 1% were found to develop disease. (Behr, Hopewell et al. 1998)

However there are difficulties in contact tracing. First, it is not straightforward to assume that contacts were infected by the identified source case and carry the same strain. In a San Francisco study, 30% of those contacts with disease were found to carry a distinct strain. (Behr, Hopewell et al. 1998) Contacts of smear positive cases may have other risk factors for disease. In particular, both the source case and contact may be within a high risk group. In addition, contact tracing has limitations in detection. In another San Francisco study, in 70% of clusters contact tracing failed to identify secondary cases. Although this was within the context of well functioning control program, it was considered that delays in diagnosis of the source case and evaluation of contacts contributed to missing cases. (Chin, DeRiemer et al. 1998) Other studies have also found that even very good control programs which include contact tracing for active detection often miss cases. Even under the best programs,

there are often not enough resources to thoroughly investigate contacts. (Reichler, Taylor et al. 2002; Reichler, Reves et al. 2002)

In order to optimize available resources for contact investigation, resources may be focused on those at highest risk. Several studies have developed models to examine the usefulness of contact investigations. (Weis 2002) (Gerald, Tang et al. 2002)

Depending upon local control priorities, contact tracing can be used to the appropriate extent and for either detection of new cases or infections. In high burden developing countries, the priorities may need to focus on identifying new cases that may otherwise not have received treatment. In low incidence regions, contact investigation is unlikely to detect many new cases, and the focus can turn to screening contacts for TB infection and treatment to prevent progression to disease. (Wares, Akhtar et al. 2000; Pronyk, Joshi et al. 2001)

VII. B. Screening amongst high risk groups

In low incidence regions, there emerge sub-populations that are at higher risk for disease. These often include immigrants from high-incidence countries, close contacts of infectious cases, homeless, drug users, and those co-infected with HIV. Active screening amongst targeted high-risk groups may be beneficial. A high proportion of cases occur amongst the poor, who may have less access to health services. Screening programs connected with homeless shelters and drug clinics may identify cases which otherwise would remain untreated, and reduce cycles of transmission amongst groups sharing confined spaces. Active screening within high-risk groups should include skin testing for diagnosis of latent tuberculosis infection, followed by preventive treatment of LTBI. Chest x-rays for diagnosis of active disease is not recommended as a screening tool amongst non-symptomatic individuals in high-

risk groups. Rather, chest x-rays are reserved as a diagnostic tool. (CDC 1998; American Thoracic Society 2000; Society 2000)

Screening of new immigrants has become increasingly important in TB control in low incidence countries. As the incidence amongst the native population declines in developing countries, an increasing number and proportion of cases are found amongst immigrant populations. Immigrants from regions of high incidence have a high probability of latent infection and risk of progressing to disease soon after arrival. (Nelson and Fingar 1995; CDC 1998; Lobato and Hopewell 1998; Dasgupta, Schwartzman et al. 2000)

Estimates from the CDC suggest that there are at least 7 million foreign-born persons in the United States infected with TB, and that 2-3% of these will develop disease unless given preventative treatment. An estimated 30% of cases occur in foreign-born. (McKenna, McCray et al. 1995) Difficulties in screening and providing treatment to foreign-born individuals include the size of the immigrant populations, access, cultural barriers, and previous BCG vaccination or exposure to environmental mycobacteria. (CDC 1998; Lobato and Hopewell 1998; American Thoracic Society 2000)

VII. C. Active case detection in low incidence regions, amongst high-risk groups

Active case detection can play an important role in TB control in low incidence settings of developed countries. An important component of active detection programs is contact tracing. Effective programs include the identification of contacts of infectious cases, followed by skin testing and preventive therapy, and diagnostic (smear, culture, or chest x-ray) testing of contacts at high risk for or showing symptoms of disease. (Society 2000)

Studies have also considered the relative effectiveness of screening all contacts of an index case versus targeted screening of those at high risk. Much discussion has focused on attempting to define factors that put contacts at high risk for infection. The first obvious risk factor is close contact, but again there is difficulty in defining what constitutes close contact, and how much exposure to an infectious case puts an individual at significant risk of infection. (Marks, Taylor et al. 2000; Bailey, Gerald et al. 2002; Broekmans, Migliori et al. 2002)

In the context of a low incidence environment, studies have suggested that screening of casual or social contacts has limited benefits. Studies have suggested that focusing on screening of close contacts is more cost-effective than expanding investigations to include casual contacts, which identify few additional infections or cases. (Ansari, Thomas et al. 1998; Marks, Taylor et al. 2000)

A second important component of active detection is screening within high-risk groups. In a low incidence setting, TB cases will increasingly be found clustered within groups at greater risk for infection or disease than the rest of the population. High-risk groups include new immigrants from countries with high incidence, poor or homeless, drug users, and AIDS patients.

Screening programs have been implemented in a variety of settings in which there is a high risk of transmission amongst effectively close contacts outside the household setting. For example, case finding in prisons may be an effective means of detection and controlling of an outbreak. (Brock, Reeves et al. 1998; Saunders, Olive et al. 2001) Screening within hospital or emergency rooms can also be important in stopping nosociomal transmission cycles. (Asch, Leake et al. 1998)

One of high-risk factors for TB disease is poverty or homelessness. Screening amongst these populations, which have less medical access, can be important in identification of new cases which would be unlikely to be found through traditional

passive detection. (Southern, Premaratne et al. 1999; Moss, Hahn et al. 2000; Solsona, Cayla et al. 2001) In some settings, screening within emergency departments has been more effective than screening in homeless shelters, for identifying new cases amongst homeless. (Nelson and Fingar 1995; Asch, Leake et al. 1998)

In addition, preliminary models have considered cost-effective means for screening contacts. A model for targeted contact investigation suggests the importance of identifying risk-factors in contacts, and recommends testing only those contacts at highest risk of infection. (Bailey, Gerald et al. 2002) Other cost effectiveness studies of active screening programs have suggested that these are not always cost effective. In order to remain cost-effective, active screening programs need to rigorously keep to guidelines, identify a high proportion of contacts, provide skin testing to close contacts, and maintain high levels of preventive therapy amongst those identified with latent infections. Programs tend to be more cost-effective when there is higher risks of disease and rate of screening. (Macintyre, Plant et al. 2000) While screening of close contacts is often shown to be beneficial in identifying new infections and preventing new cases, screening new of new immigrants is not always cost-effective. (Dasgupta, Schwartzman et al. 2000) (Dasgupta, Schwartzman et al. 2000)

VIII. Treatment Delays in detection and diagnosis

Delays in treatment of tuberculosis may occur because of either patient or medical factors. Patient delays in reporting to health authorities may occur because of lack of access, cultural or social barriers, or stigma attached to infection with tuberculosis or other diseases. Once a patient has reported to a health authority with symptoms, further delays may occur before the case is accurately diagnosed. Delays relating to case diagnosis can be caused by failure to recognize symptoms or misdiagnosis.

These may all be complicated further when patients suffer multiple ailments. In addition, once symptoms of active TB disease are suspected, most programs recommend confirmation of disease by one or more laboratory methods. Typically this includes smear and culture tests and, when needed, chest x-ray for final confirmation of active pulmonary disease. Delays may occur in scheduling time to collect needed sputum samples from the patient, sometimes requiring repeat visits or multiple samples. Repeat tests may also be needed to confirm results of uncertain tests. Individuals who find it difficult to visit a clinic may not be willing or able to return for multiple visits, even to hear the results of their tests and begin appropriate treatment. This may become particularly important in amongst the poor or homeless, and when stigma is associated with visiting clinics.

Delays may also occur in returning the results of these tests. A well-functioning laboratory service can typically return the results of a smear test within several days, and a liquid-based radiometric culture test within two weeks, and a solid-based culture test within 3 – 6 weeks. In addition, the local health authority may not have the required mycobacterial laboratory and need to send the sample to a remote laboratory, taking additional time and possible delays. Poor organization, shortages in staff or equipment, and poor technical training may all contribute to further delays as well as misdiagnosis.

The time taken from onset of case symptoms to reporting and the start of treatment vary distinctively by region. Several studies have examined factors influencing patient reporting, as well as delays in case diagnosis. Many of these factors are cultural, such as associated stigma with visiting clinics, or economical, influencing access to health services. (Needham, Foster et al. 2001) Studies have also found that delays in patient reporting were related to poor health information, and lack of information regarding the symptoms and treatment of tuberculosis. These patient

delays were found to be longer in rural areas, in patients with lower education levels, and for patients who did not have prior information regarding tuberculosis. (Salaniponi, Harries et al. 2000; Wandwalo and Morkve 2000; Lienhardt, Rowley et al. 2001; Needham, Foster et al. 2001) Improving general awareness on the symptoms of tuberculosis, available treatment, and how to access health care services are important measures to both increase case detection rates and reduce the delay in patient reporting.

Even under strong control programs in developed countries, there may be significant patient delays in seeking care. Studies examining the reasons patients delay in reporting include concern over cost of treatment, difficulties in access to health services, misunderstanding of tuberculosis, and fearing immigration authorities. Patient delay was correlated to unemployment, uncertainty about where to seek care, and a belief that they could treat themselves. Conversely, the severity of the illness and symptoms were not seen to significantly affect patient delay. (Asch, Leake et al. 1998) These findings suggest a lack of equity in treatment, such that groups with lower education and knowledge about seeking health care and tuberculosis disease itself are less likely to receive treatment.

Patients may experience delays once in hospital, waiting either for diagnosis or to start treatment. These delays may include intervals for referral, diagnosis, and initiation of treatment. Both doctor and clinic delays have been shown to contribute to overall institutional delays. (Salaniponi, Harries et al. 2000; Yilmaz, Boga et al. 2001) Clinical delays in treatment are often associated with atypical presentation, lack of symptoms, and waiting for culture results. Older patients and those without respiratory symptoms are more often initially misdiagnosed. (Mathur, Sacks et al. 1994)

Delays in treatment are associated with admittance to a hospital with a low incidence of tuberculosis, and atypical clinical presentation. In hospitals with low TB admission rates, cases were more likely to be initially misdiagnosed. Further, the rate

of TB admissions was inversely proportional to the successful treatment outcome. At lower rates of TB admissions, cases were more likely to fail treatment, die, and transmit infection. Treatment delays amongst patients in a hospital may lead to nosociomal transmission. (Greenaway, Menzies et al. 2002) Within emergency room departments there have been more specific analysis regarding treatment delays. The time to diagnosis and initiation of treatment are lower in emergency rooms and hospitals which have a higher incidence of tuberculosis cases, and are more familiar with methods for diagnosis and treatment of tuberculosis. In addition, the time from admission of an infectious case to isolation is important in preventing nosociomal transmission, and is an additional measure important in these studies. (Mathur, Sacks et al. 1994; Moran, Fuchs et al. 1995; Moran, McCabe et al. 1995)

The waiting times between onset of patient symptoms and reporting (patient delay) as well as between patient reporting and the initiation of treatment (medical delay) vary considerably by region and medical service. Examples of patient and medical delays have been estimated a wide range of settings. Studies on waiting times found in the literature are summarized in the **Table 3-1**. In general, longer delays were observed in rural as opposed to urban settings. This is likely attributable to difficulty in accessing and fewer medical resources. Longer delays were also associated with poor education, low income, and lack of knowledge about TB disease and care.

Setting	Total mean delay	Mean patient delay	Mean medical delay	Notes	Reference
Canada			1 week or more in 30%		(Greenaway, Menzies et al. 2002)
Ethiopia		60 days (median), 78.2 (mean)	6 days (median), 9.5 days (mean)	Longer delays associated with far distance to health service and poor knowledge about TB treatment.	(Demissie, Lindtjorn et al. 2002)
Zambia	9 weeks	Delay associated with female sex, outpatient diagnosis, lower education, visiting private doctor or traditional			(Needham, Foster et al.

		healer.			2001)
The Gambia, West Africa	Median delay from onset of symptoms to commencement of treatment of 8.6 weeks (range 5-17).			Delay longer in rural (12 weeks) vs. urban (8 weeks). Delay not related to cure rate, but increased death rate.	(Lienhardt, Rowley et al. 2001)
Botswana					(Creek, Lockman et al. 2000)
Malawi	8 weeks	7 weeks	1 week (smear test in 4 days)		(Salaniponi, Harries et al. 2000)
Tanzania	185 days	162 days (90%)	23 days (10%)	Longer delay in rural, amongst low education, those visiting traditional healers.	(Wandwalo and Morkve 2000)
LA, California, USA		60 days (mean 74 days, SD 216 days)		Self-reported access barriers. 1/5 patients delayed reporting for longer than 60 days. Delays more common in unemployed, those concerned with cost, difficulties seeking treatment, unsure where to get care, belief in efficacy of self-treatment, and those fearing immigration authorities.	(Asch, Leake et al. 1998)
Ghana, West Africa	4 months (median), 7.7 months (mean)	4 weeks (median)	8 weeks (median)	In 44%, delay longer than 6 months, strongly associated with rural residence. Long doctor delays associated with no sputum test, low diagnosis rates, rural setting, female patients, and those needing hospital admissions.	(Lawn, Afful et al. 1998)
Kuala Lumpur, Malaysia	12.5 weeks	7 weeks	2 weeks	Sputum tests and chest x-rays underutilized; private practitioners should be more alert to symptoms and diagnosis.	(Liam and Tang 1997)
Victoria, Australia	31% of patients started treatment within 31 days of symptoms onset. 86% of smear positive cases started treatment within 3 days after positive smear test; longer medical delays were only found in private treatment.				(Pirkis, Speed et al. 1996)

Table 3-1 ***Treatment waiting times and delays.*** The delays in case treatment reported in studies listed below are summarized according to region. The treatment delays are reported as the total mean delay from suspected time of symptoms onset to initiation of treatment. Some studies differentiated further as the patient and medical delays. The patient delay represents the time from onset to patient reporting to health services. The medical delay refers to the time from patient reporting to initiation of treatment

IX. Case Diagnosis –methods

Cases of active TB disease are diagnosed by a variety of means, including traditional smear and culture methods, as well as newer protocols such as radiometric and molecular methods. All of these tests are based upon obtaining and processing sputum samples. Sputum samples are collected from patients with persistent cough that produces secretions from the lower respiratory tract. Sputum specimens should be refrigerated and rapidly conveyed to the laboratory for tests. Sputum samples are first processed for homogenization and decontamination to remove respiratory secretions and treated with basic solutions to retard the growth of other bacteria. These solutions are then removed spinning the sample in a refrigerated centrifuge, and the mycobacteria concentrated in the pellet. The pellet can then be sampled directly for presence of mycobacteria by a smear test. In order to culture, the pellet should be resuspended in appropriate media to redistribute the mycobacteria. (Lutwick 1995; Davies 1998; Iseman 2000)

In a smear test, a sample taken from the patient's sputum is smeared on a slide, stained, and examined by microscopy to determine the presence of acid fast bacilli (AFB). When performing smear microscopy, these samples isolated from a patient are treated with a dye that stains the waxy lipid-rich mycobacterial cell wall. The dye is taken up through the lipid-rich mycobacterial cell walls through heating and alkaline treatment. Once the dye is absorbed into the mycobacterial cell walls, the cells retain this stain even when treated with an acid solution. The sample is then counterstained with a dye of another colour in order to reveal the mycobacteria present on the background of other cells. (Lutwick 1995; Davies 1998; Iseman 2000)

Smear microscopy is the most commonly used method for diagnosis of active disease. The benefits of this method include a faster return of results, but this is balanced by the difficulties in diagnosing tuberculosis when the patient is infected with nontuberculosis mycobacteria. While the smear test has lower specificity and

sensitivity than culture techniques, the low cost, the ease of administration and interpretation have allowed the smear test to remain the most widespread method for diagnosis. In particular, in many resource poor settings this remains the only available means of microbiological confirmation of active TB disease. Most cases with cavitary lung disease are strongly smear positive, while patients with non-cavitary disease are likely to be smear negative. The sensitivity of the test appears to be able to detect at a level of above 7,800-9,500 bacilli per mL of sputum. (Yeager, Lacy et al. 1967; Hobby, Holman et al. 1973) The test is also limited by specificity; other non-tuberculosis mycobacteria also have the same acid-fast properties that allow for the stain to be retained and show a positive smear result. The usefulness of smear in diagnosis is therefore also related to the prevalence of non-tuberculosis mycobacteria in the environment, and the proportion this contributes to mycobacterial disease. (Lutwick 1995; Davies 1998; Iseman 2000) (Perkins and Kritski 2002) (American Thoracic Society 1990)

In culture-based diagnosis, a sputum sample isolated from the patient, as described above, is grown either in liquid or on solid medium. The choice of medium will affect the growth time required to detect the presence of mycobacterium. When grown on solid medium, the positive detection of mycobacterium takes from 3 to 8 weeks. Culture-based tests have high sensitivity and specificity. Further, these tests are relatively both affordable and are easily performed, making them well suited for the developing world. (Lutwick 1995; Davies 1998; Iseman 2000) (Hale, Pfyffer et al. 2001; Perkins and Kritski 2002)

Use of liquid based media and radiometric techniques, such as the automated BACTEC system, can reduce the detection time to about two weeks. (Middlebrook, Reggiardo et al. 1977) In repeated trials, the BACTEC system has been shown to recover over 90% of *M. tuberculosis* specimens. (Roggenkamp, Hornef et al. 1999;

Harris, Rayner et al. 2000) Systems similar to BACTEC have been developed that allow for rapid and accurate detection of mycobacterium without the need for radioactivity. The most commonly used alternative is the automated non-radiometric MB/BacT system. (Yew, Tong et al. 2001) The MB/BacT system has been shown to be reliable for detection of mycobacterium in culture. In some trials, the MB/BacT system has shown slightly lower sensitivity in detection of mycobacterium than the more traditional BACTEC system, in particular for detection of smear negative cases. (Roggenkamp, Hornef et al. 1999; Harris, Rayner et al. 2000; Watterson and Drobniewski 2000)

There have also been efforts to develop routine serological tests for tuberculosis, particularly for diagnosis of smear negative pulmonary disease. Such tests have been developed to detected antigens on the surface of the tubercle complex. However, these methods have largely lacked sensitivity in detection, as well as specificity due to cross reaction with other mycobacterial species. The use of ELISA tests for direct detection of antigens on *M. tuberculosis* have been developed from murine monoclonal antibodies. (Watterson and Drobniewski 2000) (Lutwick 1995; Davies 1998; Iseman 2000)

In addition, tests have recently been developed for direct detection of disease through PCR. Such tests can reduce the diagnostic time from several weeks to several days. In addition, molecular amplification tests offer high specificity and sensitivity. In these tests, sputum samples from patients are treated to isolate DNA, which is then amplified by PCR for either sequencing or hybridization and RFLP analysis. PCR-based tests have become commonly used for determination of mycobacterial species in some regions, with two direct *M. tuberculosis* nucleic acid amplification tests approved by the Food and Drug Administration in the USA. DNA microarrays have also been

used to rapidly screen for multiple mycobacterial species. (Soini and Musser 2001) (Watterson and Drobniewski 2000) (Hale, Pfyffer et al. 2001)

X. Drug testing—methods

X. A. Need for standardized protocols in network of centralized laboratories

Drug testing is needed both for regional surveillance and for individual patient treatment. Drug resistance has existed since chemotherapy of tuberculosis began. Over the past decade, resistance has been found in every region surveyed and epidemics of MDR have placed an increasing burden on TB programs. (WHO/IUATLD 1997; WHO/IUATLD 2000) In order to prepare for the surveillance presented by the WHO working group on drug resistance surveillance, a network of laboratories was created in participating countries that coordinated case detection and drug testing methods. High standards of drug testing were maintained in these laboratories, although the methods varied slightly according to available resources in particular laboratories. The coordination of efforts allowed for the comparison of these methods across the laboratories in the network. Overall, these drug susceptibility testing methods showed a high degree of specificity and sensitivity, although the sensitivity tended to be a bit lower. Across the laboratories in the network, there was a high degree of concordance in the sensitivity of drug testing for isoniazid and rifampicin, but disagreement for susceptibility testing of other first line drugs. (Laszlo, Rahman et al. 1997)

Recommendations for surveillance of resistance in Europe recommends establishing a network of laboratories that coordinate methods in order to ensure high proficiency and quality of testing, as well as the ability to compare results for regional surveillance. The working group further recommended that when resources are available, there should be drug susceptibility testing of all isolates. Drug testing for

resistance to at least isoniazid and rifampicin should be recorded, and for surveillance purposes, linked to records of case notification, diagnostic tests, and critical case information. (Schwoebel, Lambregts-van Weezenbeek et al. 2000)

Drug susceptibility testing is also needed to guide patient treatment, allowing for prescription of optimal regimens by case. In order to ensure proficiency in and optimization of drug susceptibility testing methods, it is important to maintain high proficiency in laboratories performing these tests. Because maintaining such technical expertise, as well as obtaining all necessary resources, may be difficult within local laboratories, it is important that samples can be sent to regional laboratories. Such central laboratories can maintain the expertise needed. In addition, localization of needed resources within central laboratories can provide the needed drug testing at lower costs. (Heifets and Cangelosi 1999)

X. B. Culture based methods

The traditional method for diagnosing patient resistance to particular tuberculosis drugs is based on culture. A case sputum sample, containing active mycobacterium, is cultured on either solid or liquid medium containing that particular drug. Growth in the presence of drug is considered to show resistance. Typically, the sample is cultured on a number of different dilutions of drug, to allow the relative comparison of growth, and estimation of the minimum inhibitory concentration (MIC). The choice of medium will affect the time taken to culture the sample to detectable levels, but on average this takes 3 to 5 weeks. Sensitivity and contamination are also issues with culture based drug testing. Contamination of cultures may lead to false positives for resistance.

There are three basic methods for measuring resistance in culture. In addition, for any of these three methods, resistance can be tested for either directly, taken from

the smear positive sputum, or indirectly, taken from colonies cultured in laboratory. While direct testing may allow for more accurate representation of the proportion of resistant organisms present in the sputum sample, indirect testing allows for selection of viable mycobacterium. The absolute concentration method involves inoculating a sample on plates containing varying concentrations of drug, as well as a control plate with no drug. If more than 1% of the inoculum grows on media containing critical concentrations of the drug, the sample is considered to be resistant. The resistance ratio method compares the growth of a patient sample to a standard laboratory strain. The results are reported as the ratio of the MIC of the patient to laboratory strain. The proportion method compares the growth of a strain in the presence and absence of drug. The sample is considered resistant if more than 1% of the colonies present on the control grow on media containing drug at a critical concentration.

X. C. Automated radiometric methods

With the increasing use of automated methods for detection of mycobacterium, such as the radiometric BACTEC system, methods have also been adapted to allow use of these automated systems for detection of resistance. The BACTEC system has been used allow for faster detection of resistance by the proportion method. The growth of patients sample is compared in the presence of drug and a 1:100 dilution of the sample in the absence of drug. The growth index values from two consecutive days are compared; the sample is considered to be resistant if there is slower growth in the sample with drug compared to the control. (Roberts, Goodman et al. 1983; Lee and Heifets 1987) (Rastogi, Labrousse et al. 1996; Davies 1998; Iseman 2000)

The radiometric BACTEC method has been used to determine resistance is patient samples. The initial method took several weeks for detection of growth of resistant organisms. (Roberts, Goodman et al. 1983) (Siddiqi, Hawkins et al. 1985)

The MIC of strains has also been determined using the BACTEC method. The MIC as determined by BACTEC is considered to be closer to the true MIC of the strain present in the patient than as cultured on agar plates, as this method is faster, allows for more absorption and is more quantitative. (Lee and Heifets 1987) (Rastogi, Labrousse et al. 1996) A multicenter trail using Bactec reported susceptibility results completed in 11-23 days, compared with 33-42 days for conventional tests. (Middlebrook, Reggiardo et al. 1977) In more recent studies, use of the BACTEC method for susceptibility testing has been completed in an average of 8 days. (Pfyffer, Bonato et al. 1999)

A more recent comparison of resistance tests by the automated, nonradiomateric MB/BacT system and the agar proportion method found the MB/BacT system to be as reliable as culturing on agar. The MB/BacT system was also considerably faster; the mean time to using the MB/BacT system was 4.7 days for resistant isolates and 10.8 days for drug sensitive isolates, compared to 14.8 days for resistant and 21 days for sensitive isolates on agar plates. (Yew, Tong et al. 2001)

X. D. Sensitivity and reliability of DST

While this test does take the longest time to return results, it is affordable even in the developing world, and requires only relatively simple technical skills. However, the reliability of these tests is still dependent upon lab techniques and in particular attempts to avoid contamination. A further difficulty with culture-based resistance tests is that they can signal the presence of resistant mycobacteria in the sample, but not the relative amounts of sensitive and resistant organisms.

X. E. Molecular tests for resistance

Diagnostic tests relying on molecular methods use knowledge about the genetic basis of resistance to a particular drug. About 40 identified mutations or deletions to date have been shown associated with a resistance phenotype. The genetic tests are

most developed for the identification of rifampicin resistance, for which greater than 95% of mutations can be found in the rpoB gene. (Kim, Lee et al. 2001) PCR based techniques involve amplification of a region of a gene, followed by either RFLP analysis or sequencing to differentiate wild type from mutant sequences. (Soini and Musser 2001) (Caws and Drobniewski 2001; Fluit, Visser et al. 2001) Direct sequencing of the resistance gene allows for the highest detection rate of the presence of genetic resistance in a sample, followed by RFLP analysis. The usefulness of either method varies by locus and test for each drug. (Nachamkin, Kang et al. 1997)

A significant limitation of these molecular methods for resistance testing is that they do not reflect on level of resistance to a drug, and thus MICs cannot be determined from these tests. In addition, these tests only show genetic resistance, but this does not always translate into phenotypic resistance. PCR tests can typically be completed in 2-3 days time under good laboratory condition ns. It may take several weeks, however, to take a sample, send to the laboratory, and to return results. In addition, there may be need for repeated drug tests requiring further patient samples. (Fluit, Visser et al. 2001; Soini and Musser 2001; Victor, van Helden et al. 2002)

X. F. Other susceptibility tests for developing world use

There has been an emphasis recently on the use and development of molecular based susceptibility testing methods, which will allow for faster and more accurate detection of the genetic resistance. However, the use of such molecular methods is only realistic within the context of the high market economies, and the small proportion of cases which fall under well funded TB programs which can afford to allocate resources for drug testing of all cases. (Fisher 2002) The vast majority of cases are found under the coverage of under funded programs which cannot afford any resources for general case drug testing, much less the building of standardized laboratories and

technical expertise required for the newer molecular based methods. There remains a great need, as resistance rates grow within these resource-limited programs, to diagnose case resistance profiles in a timely fashion before or shortly after beginning treatment. In order to meet this need, drug susceptibility testing methods which are applicable for wide scale application in developing world settings need to be considered; such tests need to be affordable, require few additional resources and low technical skill.

A number of other tests for diagnosing resistance have been developed, and considered for their applicability to wide scale use in developing country and resource poor settings. For example, a swab test was used, and found to be as effective, cheaper, and easier to implement than indirect inoculation and culture to test media. However, this requires an average of eight weeks, the same as for traditional culture methods, to diagnose resistance. (Mathew, Nair et al. 2000) Another example is the microscopic observation broth-drug susceptibility assay (MODS), which allows for early detection of mycobacterium growth in liquid medium. MODS can be used for more rapid diagnosis of disease as well as detection of resistance. MODS offers a rapid, relatively inexpensive, sensitive and specific method for susceptibility testing. (Caviedes, Lee et al. 2000)

X. G. Global drug testing

The global use of drug testing and second line regimens are difficult to quantify. The best current estimates of prevalence of resistant cases come from WHO drug resistance surveillance project, which has been ongoing for the past eight years. This study has followed the prevalence of resistance to the major TB drugs in 54 countries/regions, by sampling a proportion of active TB cases. Case samples are sent for drug sensitivity testing, with methods standardized through a system of shared protocols within a network of coordinated laboratories. (WHO/IUATLD 2000)

In some regions, drug testing has become an integral component of the TB control program. In the low incidence regions where TB has nearly entered the elimination phase and resources are available for individualized testing and treatment, programs have recommended drug testing of all new cases. In particular, this is important in regions with a high prevalence of primary resistance, or amongst immigrants from such regions. (Clancy, Rieder et al. 1991; CDC 1998; CDC 1998; Schwoebel, Lambregts-van Weezenbeek et al. 2000; Broekmans, Migliori et al. 2002) In the UK, the PHLS gathers samples of all new cases and tests for drug resistance. (Kumar, Watson et al. 1997; Eltringham and Drobniewski 1998) Similarly in the USA, regulations recommend the drug testing of new and previously treated cases. (CDC 1998)

Outside of these programs, drug testing of TB cases is more difficult to quantify. Much of this happens in the private sector and is not officially monitored by surveillance or control programs. Some occurs within the structure of national and inter-national control programs, including DOTS-plus program. Some programs explicitly state the need for drug testing of all new cases in regions of high resistance prevalence. Other guidelines suggest that only chronic infectious cases not responding to treatment and cases with contacts of resistant patients should receive drug testing. Still, much of drug testing and treatment of resistant cases occurs in the private sector, outside of organized health authorities, and without coordinated patient records.

X. H. Treatment of drug resistant cases

Since the establishment of short course chemotherapy regimens consisting of four or five common first line drugs, acquired resistance emerges at lower rates per treated case. However, resistance to multiple drugs, particularly to INH and RIF, two of the most potent and effective first line drugs, has begun to be commonly observed.

(WHO/IUATLD 1997; WHO/IUATLD 2000) Primary resistance is a leading cause of treatment failure on first line SCC. As MDR emerged and caused noticeable strain on some health systems, due to the cost and difficulty associated with treating these cases, more studies began to focus on epidemiology and treatment of resistance. In order to treat cases resistant to one or more of the first line drugs, specialized regimens were developed. Some TB control programs developed clinical guidelines for treatment of resistant, and in particular, MDR cases. These guidelines include drug testing of new cases in order to identify resistance patterns and to modify treatment regimens accordingly. A central recommendation for treatment of resistant cases is to identify resistance early and ensure that the regimen used does not allow for monotherapy. Second-line regimens used to treat resistance provide a more effective cure than continued treatment with first-line regimens. This shift also ideally removes a high selective pressure for acquired resistance, which may occur under continued treatment with drugs to which the patient has already developed resistance. (WHO 1997; Espinal, Dye et al. 1999; WHO 1999; Bastian, Rigouts et al. 2000; Farmer 2001)

Early clinical trials examined the impact of initial resistance on treatment outcome. The results of pretreatment drug susceptibility results were correlated to outcome. Initial resistance to a single drug was correlated with slightly reduced cure rates. (Hong Kong Tuberculosis Treatment Service/ British Medical Research Council 1972) All the regimens appeared to be effective at treating cases with initial resistance to either isoniazid or streptomycin. However, the treatment was less effective with there was initial resistance to rifampicin. In addition, when neither rifampicin nor pyrazinamide were included in the continuation phase of treatment, the relapse rate for cases with initial isoniazid resistance was increased. (Hong Kong Tuberculosis Treatment Service/ British Medical Research Council 1972) (Davies 1998; Iseman 2000) In another trial, the role of primary resistance on response to SCC was examined

again within 12 control trials in Africa, Hong Kong and Singapore. Amongst cases with initial resistance to isoniazid or streptomycin, 17% of those given a 6 month regimen of isoniazid and rifampicin, and 12% of those given rifampicin in the initial 2 month phase failed treatment. The proportion of treatment failures decreased with increasing number of drugs used in the regimen, and a longer duration of treatment with rifampicin. The treatment failure rate was only 2% for cases which received a 4 or 5 drug regimen that included rifampicin throughout the 6 months of treatment. (Mitchison and Nunn 1986)

More recently, clinical trials have shown that MDR cases have poor response on SCC. MDR is the major cause of treatment failure on the DOTS retreatment regimens. (WHO 1997) Patients with patterns of MDR resistance showed low cure rates on the first line regimen. In addition, treating patients initially resistant to one or more drugs with these first-line drugs could allow for some periods of effective monotherapy, during which there could be rapid selection for resistance to the remaining drugs. Many patients developed resistance to all of the traditional drugs, and again, some turned to alternative treatments such as surgery. More recently, the development of new drugs, including various second-line drugs have been introduced in treatment of these multi-resistant cases. While many of these drugs remain early in use and are not as well characterized as the first line drugs, they have shown effectiveness in treatment of resistant cases. (WHO 1997) (WHO 1997; Espinal, Dye et al. 1999; WHO 1999; Bastian, Rigouts et al. 2000; Farmer 2001; Sterling, Lehmann et al. 2003)

In response to the poor response of MDR cases to SCC, specialized regimens were developed to treat cases with initial resistance. The recommended regimens for treatment of MDR cases includes an initial intensive phase lasting at least 6 months, followed by a continuation phase of 12 to 18 months. During the initial phase, 4 or

more drugs should be used, and 3 or more of the most active drugs should be used in the continuation phase. (WHO 1997; WHO 2003)

Resistance can either be treated using standardized or individualized regimens. Where resources for drug testing are available, all cases failing treatment should receive drug testing. Cases diagnosed with resistance should then be placed on specialized regimens. When resources for drug testing are not available, standardized regimens should be used for retreatment. However, cases failing retreatment regimens should be referred to either specialized centers for resistance treatment or receive retreatment regimens that are based on knowledge of the likely resistance patterns. There are several benefits to using regimens standardized within the region for treatment of MDR cases. First, this allows for a lower cost (WHO 1997; WHO 1999; Bastian, Rigouts et al. 2000; Farmer 2001)

Programs can also offer individualized treatment for MDR cases through centers of excellence. Referring MDR cases to specialized centres allow individualized treatment with less effort and changes to local care practices. Such centres can achieve high cure rates for MDR cases with lower cost, by offering individualized treatment regimens tailored to drug susceptibility patterns of the patient, can give fully observed therapy and ensure adherence through a long and difficult treatment. However, such care is costly, including expensive second line drugs, testing facilities, laboratories, and a high level of care. (Farmer 2001; Mitnick, Bayona et al. 2003; WHO 2003).

XI. Summary

In this chapter, I review literature on the treatment of TB. I begin with a historical perspective, starting with the introduction of the early chemotherapy regiments in the 1960s. While these were typically based on monotherapy, they did offer a rapid cure in a high proportion of cases. As was soon realized, however,

monotherapy led to high levels of drug resistant cases. Introduction of rifampicin, which had effective sterilizing activity, in the regimens in the later 1960s vastly improved cure rates. Today, the standard chemotherapy regimens are based on the World Health Organization's DOTS-SCC program. The basic requirements of DOTS programs are briefly reviewed here. I also provide an overview of what constitute the basic elements of a control program, including the case detection and cure rates, and survey these for TB programs. I also review the current standards and methods for case diagnosis and drug susceptibility testing. This review provides basis to understand the context of current treatment practices for TB and control programs, and to understand the development of the model in Chapter 5 for TB transmission, which includes treatment, as well as the model developed in Chapter 8 that includes many of the elements of control programs discussed here.

Chapter 4

Review of mathematical models of tuberculosis epidemiology

"It was the fashion to suffer from the lungs; everybody was consumptive, poets especially; it was good form to spit blood after each emotion that was at all sensational and to die before reaching the age of thirty"

-- Alexander Dumas

CHAPTER 4

REVIEW OF MATHEMATICAL MODELS OF TUBERCULOSIS EPIDEMIOLOGY

I. GOALS

II. ABSTRACT

HI. INTRODUCTION

IV. INTRINSIC DYNAMICS OF EPIDEMICS

IV. A. DISEASE RISKS
IV. B. LONG TERM DYNAMICS OF EPIDEMICS
IV. C. TB IN ENGLAND AND WALES

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VI. DRUG RESISTANCE

VI. A. EMERGENCE OF ACQUIRED DRUG RESISTANCE
VI. B. MDR TRANSMISSION (AND EVALUATING EFFECTIVENESS OF CONTROL STRATEGIES)

VH. CONCLUSION

I. Goals

I. A. To review the literature on mathematical models of tuberculosis epidemics.

II. Abstract

In this section, previous mathematical models of tuberculosis are reviewed. The models and studies included have made critical contributions to understanding historical tuberculosis epidemics and their intrinsic dynamics. Models which focus

specifically on understanding the dynamics of drug resistant tuberculosis are also examined. The critical components of these models are compared in Table 4-1.

III. Introduction

Mathematical models for infectious diseases are used for many purposes within the study of infectious disease epidemiology. Epidemiological models may focus on the transmission and dynamics of an epidemic within a population. These models may also include control strategies, and be used to weigh the benefits of possible interventions. Other models may consider the dynamics of a bacterial or viral infection within an individual. Such within-host models may be used to consider the impact of drug treatment on the infection, the interaction with the host immune system, and the emergence of drug resistant organisms. In addition to the range of possible uses of mathematical models in the study of infectious diseases, the methods used in developing the model are also important. The types of models typically used are deterministic, defined by a set of ordinary or partial differential equations, or stochastic. Models may consider only the disease state in subdividing a population, or may further subdivide according to categories such as age. The inclusion of additional categories should be appropriate and justified by the disease and situation. Ideally, both the methods and type of models will be selected as those best suited to the question at hand.

In this chapter, the literature on mathematical models of TB is surveyed, highlighting those mainstream and influential studies most relevant to this thesis. First, models used to understand the natural history and epidemiology of TB are reviewed. Second, models used to consider various treatment strategies for the control of TB are presented, followed by a final section on models that include drug resistance. The most

relevant elements of each of these models, including parameters for basic disease as well as control, and for model formulation and design, are compared in Table 4-1.

Many of these elements (shown as 'Bahcall' in Table 4-1) are considered in the models developed in later chapters of this thesis. As shown in Chapter 6, when the sensitivity of the model parameters is examined, the parameters that are most central to modeling resistance are the inclusion of both acquired resistance and primary transmission. The inclusion of multiple strains within one population, is required in order to examine the dynamics for emergence of resistance from within an entirely drug sensitive epidemic (as all were pre-chemotherapy). In addition, this is needed to model the dynamics between sensitive and resistant strains that may continue after their emergence, and is likely to be central to understanding current epidemics, in which resistance has remained at relatively low prevalence.

In the chapters to follow, we develop a model to address the emergence, transmission, and control of resistant TB. This model includes many of the aspects of previous models reviewed here and considered essential to TB models. In addition, the model adds new components suited to address the questions posed in this thesis. In Table 4-1, the components of the model developed in later chapters of this thesis ("Bahcall") are compared to the models reviewed in this chapter.

Some of these studies, as noted in Table 4-1, have included age structure. Since age structure can be an important component of TB epidemiological models, I also considered the use of age structure in developing models within this thesis. However, based on in initial simulations of a model incorporating age structure, presented in Chapter 5, it is suggested that age structure has limited impact on the overall model conclusions, and is therefore not used for the subsequent model simulations throughout the thesis. Within these limited studies presented in Chapter 5, examining the prevalence of resistance by age in England and comparing these to simulations with

model that incorporates age structure, we conclude that that this does not significantly impact on model predictions for overall prevalence of resistance, dynamics between strains, or on the impact of control. However, if in considering a particular scenario, there appeared to be strong relationship between age and a particular parameter key to the models as developed here, it may prove helpful to incorporate age structure into a model. For example, if studies revealed significant variation in susceptibility to a particular resistant strain by age, this could affect resistance prevalence and the relative transmission and success of the resistant strain. Similarly, if treatment efficacy for resistant cases varied significantly by age, this could impact model predictions. However, from current studies, there has not been reason to suggest that the resistance prevalence by age will vary, or reflect more than the relative disease risks by age, combined with the relative proportion of resistant strains circulating in the population.

IV. Intrinsic dynamics of epidemics

Early models of TB spread and persistence have largely focused on an historical dynamics perspective of defined epidemics. These models have shown that the long latency period of tuberculosis results in epidemics with slow dynamics.

IV. A. Disease risks

Early models from Sutherland were used to estimate the risk of developing disease from surveys on military recruits in the Netherlands. The relative contribution of primary, endogenous, and exogenous disease to annual incidence was estimated. The highest disease risk was found to occur shortly after primary infection, followed by reinfection, and finally latent infection with no reinfection. (Sutherland, Svandova et al. 1976) The risk of developing disease within five years after primary infection was estimated at 5.06% per anum. For five years after a reinfection event, the annual risk of developing disease was slightly lower at 1.91%. In the absence of reinfection, latently

infected individuals experienced a 0.0253% annual risk of endogenous reactivation. Latent infection was also shown to confer protection from reinfection disease, estimated at 63% (for men) and 81% (for women) protection. (Sutherland, Svandova et al. 1982)

IV. B.Long term dynamics of epidemics

More recently, Blower has examined the intrinsic dynamics of long term TB epidemics. The model presented by Blower et al. consists of three states: susceptible, latent, TB disease (S, L, T), in which infected individuals can progress to an infectious case either directly (primary) or after a period of latency (reactivation). In an expanded five state model, active TB cases are either infectious (T_i) or non-infectious (T_n), move to the recovered class through natural cure, and return to disease through relapse. (Blower, McLean et al. 1995)

Using these two models, Blower demonstrates that tuberculosis epidemics can be modeled as the combination of a series of time-lagged epidemics, representing the fast dynamics of primary progression to disease and the slow dynamics of endogenous reactivation. The time lag between the rise of these sub-epidemics occurs because the dynamics of each are age-dependent with reactivation TB occurring more often in older individuals. During the course of an epidemic, the relative proportions of each sub-epidemic will shift. Fast cases (primary disease) predominate early in an epidemic, while slow cases (reactivation disease) predominate in a mature epidemic. Similarly, the age-distribution in an epidemic will shift toward older individuals as the contribution of reactivation disease increases with progression of an epidemic. Using this model, Blower et al. demonstrated the slow intrinsic dynamics of tuberculosis epidemics, with an estimated mean epidemic length of 100 years. The authors also define the basic reproductive number of each sub-epidemic, and the threshold susceptible population size required for a TB epidemic to emerge. (Blower, McLean et

al. 1995) Sensitivity analysis reveals that the parameters most significantly affecting the epidemic are equilibration time, the effective case contact rate, the fraction of new infections that develop primary disease, and the TB mortality rate. (Porco and Blower 1998)

The central conclusions of these papers by Blower et al. are that TB epidemics occur on long timescales, and can be understood in terms of the combination of fast and slow progression to disease. These are conclusions central and common to the understanding of TB epidemics, repeated and verified by a number of models.

IV. C. TB in England and Wales

Vynnycky and Fine have rigorously characterized the dynamics of the tuberculosis epidemic in England and Wales over the past century. (Vynnycky and Fine 1997) The authors described the dynamics of TB infection and disease since 1900 using an age-structured deterministic model which includes classes for susceptible, immune, infected, latent, reinfected, endogenous and exogenous disease. Infected individuals are at risk for primary disease for five years, after which time infecteds who have not developed disease move into a latent class (with a lower rate of progression to disease).

The effective contact rate is defined as contact sufficient to lead to infection, and is derived each year as the annual rate of infection divided by the prevalence of infectious cases. As this information was not available prior to 1950, trends in age-specific mortality rates were used to estimate tuberculosis prevalence during these years. They estimate that with a case fatality rate of 50% in the prechemotherapy era, the average duration of infection was 2 years. The introduction of chemotherapy in the 1950s gradually reduced case fatality to 25% and duration of infection to 1 year. (Vynnycky and Fine 1997)

Vynnycky finds that a model allowing for reinfection provides a better fit to the data. The best fit came from the assumption that reinfection occurs at the same rate as first infection, but is less likely to result in primary disease progression. This suggests that prior infection confers partial (but not complete) immunity to exogenous disease, and that reinfection contributes significantly to the long term trends in tuberculosis prevalence. (Vynnycky and Fine 1997)

V. Control strategies

Mathematical models of TB transmission dynamics have also been used to analyze the effectiveness of different interventions on the prevalence and incidence of infection and disease. The current predominant strategy is the World Health Organization (WHO) sponsored national treatment program, DOTS. Dye provides a model for DOTS control of TB, based upon an age-structured model of (drug-sensitive) TB transmission. (Dye, Garnett et al. 1998) In this model, latently infected individuals can progress to disease either by endogenous reactivation or exogenous infection. Active TB cases are either infectious (I) or non-infectious (N). Cases receive either good treatment, and return to the infected latent class, or bad treatment, leading to treatment failure. The model also provides for self-cure, smear conversion, and the immunization of susceptibles. The DOTS strategy is incorporated into the model with estimates for case detection and cure rates. This model is used to consider the impact of current and higher levels of case detection and DOTS treatment on the course of TB epidemics in global settings. Increasing case detection and DOTS cure rates are shown to reduce incidence of disease and deaths due to TB. (Dye, Garnett et al. 1998)

Murray has also produced a model considering the effectiveness of different interventions. Murray presents a more detailed model of the transmission and treatment of drug-susceptible TB, including INH chemoprophylaxis, superinfection of latently

infected individuals, and three clinical types of active TB disease (smear-positive pulmonary, smear-negative pulmonary, and extra-pulmonary). In this model, infected individuals progress to active disease at either a fast (1-5 years) or slow (5+ years) rate, and can receive INH preventive therapy, which slows the rate of progression. Infected individuals with slow latent TB experience protection from reinfection v, and are reinfected at a rate $(1-v)$, leading to fast progressive TB. Untreated cases have either a fast or slow rate of case detection and diagnosis. Treatment of infectious cases is either good (complete regimen) or bad (incomplete), with distinct cure and death rates. Recovered cases relapse to active TB at either a fast or slow rate. (Murray and Salomon 1998)

This TB transmission model is considered both in the presence and absence of HIV co-infection, making this model useful for evaluating the impact of control strategies in levels with high HIV prevalence. The authors consider the impact on incidence and mortality of supplementing current DOTS with additional strategies. Their analyses incorporate projections for the level of DOTS implementation, the extent of BCG coverage (including assumptions on duration of efficacy), and the level of HIV infection. (Murray and Salomon 1998)

VI. Drug resistance

With the increase in drug resistant tuberculosis over the past decade, models have also considered the development of drug resistance. Blower built upon her previous three compartmental model (S,L,T) to include the development of drug resistance. She presents both a model with one TB strain (DS) and with two strains (DS and DR). In the two-strain model, susceptible individuals can be infected with either a DS (S) or DR (R) strain, and either remain latently infected (L_S or L_R) or

progress directly to an infectious case (T_S or T_R). Individuals with a latent drug-sensitive infection (L_S) can also receive chemoprophylaxis and be cured. Infectious cases of both DS and DR receive treatment, but the latter is treated less effectively. Treated DS cases develop drug resistance and move into the T_R class at a constant rate. This allows for easier mathematical manipulation of the model, but does not seem an appropriate definition of acquired resistance, which should be a set proportion of treatment failures that develop resistance. New cases of acquired drug resistance are perpetuated through transmission of primary drug resistance. The possibility of a fitness cost of resistance is considered in terms of a reduced rate of transmission ($\beta_R < \beta_S$). (Blower, Small et al. 1996)

The equilibrium conditions of this model provide for three outcomes: the persistence of DR, the persistence of both DS and DR, or the eradication of both DS and DR TB. Which outcome occurs is determined by comparing the effective reproductive number for DS and DR. Also proposed is an evaluation criteria r_{MAX}, defined as the maximum acceptable probability of treatment failure, given the number of DR cases produced for each DS case. A control strategy, as specified by parameters for the effective treatment rate, the emergence of acquired resistance, and the relative treatment efficacy of DR TB, can be evaluated in terms of the effective reproductive number and r_{MAX}. (Blower, Small et al. 1996)

This analysis does contribute an understanding of the application of theoretical thresholds for model outcomes to inform on the impact of treatment. However, the application of these thresholds is limited by model structure. The idea of using the defined value r_{MAX} is both interesting and important in providing some measure for control programs to use in assessing the resistance problem. It is also nice to see a quantitative measure for the relative production of acquired resistance, as this does reflect on the state of treatment and effectiveness of the control program. The relative

production of acquired resistance will also reflect on the likely future course of the epidemic, and signal need for improved case management. However, the quantity defined in this model, r_{MAX}, may not be the most useful measurement for such considerations. First, the rate of acquired resistance is defined as a steady proportion of the treatment rate, when this should be only amongst cases failing treatment. Second, the treatment cure rates for both sensitive and resistant cases need to be incorporated. Third, the number of resistant cases produced for each sensitive case is in practice difficult to measure. A more practical alternative might include consideration of the rate of resistance amongst new and previously treated cases. Such measures can be compared to surveillance data, with the limitations as described in chapter 1 for the measurement of resistance prevalence.

VI. A. Emergence of Acquired Drug Resistance

Dye presents another model for the emergence and transmission of drug resistant TB, expanding on Blower's model by including classes for previously treated individuals. The model is used to estimate rates of acquired resistance to each drug, and to predict trends in prevalence of resistance over time.

This model includes latent classes for slow and fast progression to disease for sensitive (L_S and L_F) and resistant (L'_S and L'_F) strains, representing reactivation and primary disease, respectively. Again, there are both infectious (I) and non-infectious (N) TB disease, but only the former receives treatment. Infectious drug sensitive cases self-cure at a rate n and relapse from self-cure at a rate r_n. Infectious cases are detected and treated at a rate δ and a proportion κ of DS cases are cured. $(1-\kappa)$ of treated DS cases fail, and move into a previously treated class. Of the DS patients who fail treatment, a proportion ρ develop acquired drug resistance and move into previously treated resistant class (P'), while $(1-\rho)$ fail treatment and remain drug sensitive (P).

Infectious drug resistant cases (I) are cured at a rate κ'. The relative fitness of resistant (DR or MDR) strains in terms of the strain transmission is estimated as the relative transmission coefficient (c'/ c). (Dye and Espinal 2001)

The authors use this model to predict typical rates of acquired resistance, defined here as the probability that a patient who fails treatment will gain drug resistance. The rate of acquired resistance (ρ) is estimated for each of the four first line TB drugs. Sensitivity analysis showed that the final drug resistance prevalence is most significantly affected by the relative transmission rate of resistant strains. If a drug resistant strain is assumed to have equal transmissibility as its sensitive counterpart, then the resistant strain (with a greater R value) will in time replace all sensitive strains. However, if the resistant transmission is reduced enough so that $R_R < R_S$, then both sensitive and resistant strains will coexist. Using assumptions for the transmission rates of resistant strains, the authors predict the equilibrium prevalence of resistance, finding that resistance should saturate at 1-5% (low value for INH, high value for MDR) of all TB cases in a population. The authors note that this is approximately the current observed levels of resistance in many countries, suggesting resistance is approaching its saturation limit. The authors add a word of caution that these conclusions depend strongly on the estimates for the transmissibility of resistant strains. (Dye and Espinal 2001)

This analysis makes several important contributions. First, the rate of acquired resistance to each drug is estimated in a novel method from surveillance data. Here, the acquired resistance rate is assumed to be equal to the relative proportion of previously treated cases which are resistant. This is a useful measure for estimating the approximate rates of acquired resistance amongst treatment failures. These estimates, for other measurements of acquired resistance, are limited by estimation of rates of acquired resistance from surveillance and defining cases as new or previously treated.

Second, it is a good idea to examine the relative fitness and identify important factors in understanding the competition and transmission of resistance. These studies are also limited, as previous, by the estimation of transmission rates for resistance cases. Later studies looked at the range of resistance from estimates, using these range estimated ranges for predicted resistance prevalence. (Dye, Watt et al. 2002; Dye, Williams et al. 2002)

VI. B.MDR transmission (and evaluating effectiveness of control strategies)

Another model focuses on MDR strains and addresses the control of MDR epidemics. This model considers the dynamics of an MDR epidemic separated from other TB cases (drug sensitive or singly drug resistant) that may exist in a population. The model includes transmission of primary MDR, but not the emergence of acquired resistance. In the model scenarios, MDR cases are introduced either into a totally susceptible population (in which there are no TB infections), or a population with a proportion (z) previously infected with either drug sensitive or singly drug resistant TB. Previous infection is assumed to afford some immunity against reinfection with MDR, thus increasing the proportion of non-MDR TB in the initial population slows the spread of MDR cases once introduced. Again, MDR strains are considered to have a lower fitness relative to DS strains, incorporated into the model with a lower contact rate for MDR (c_m) than for DS (c).

This model is used by the authors to consider how to target control strategies for MDR-TB. The reproductive number for MDR is defined in the absence (R_{0M}) and presence ($R_{\theta M}$) of chemotherapy, and based on treatment data from six countries (Espinal, Kim et al. 2000), they define combinations of case detection, treatment, and cure required to eradicate a MDR epidemic (in terms of keeping $R_{0M} < 1$). (Dye and Williams 2000)

This analysis contributes the idea of using defined R values to assess control of MDR epidemics. Defining such thresholds based on model parameters allows for consideration of the conditions required to prevent or control an epidemic. However, the applications of this model to real program settings are severely limited by considering only MDR epidemics in isolation. MDR emerges through the treatment of sensitive cases, and continuing MDR transmission depends upon the dynamics and interaction with the overall TB epidemic. Because epidemics of MDR cases are considered in the absence of interaction with drug sensitive cases, the R values defined are applicable only for MDR epidemics in isolation. These R values are the same as for drug sensitive epidemics, differing only by transmission and cure rates. Thus, while it is useful to define R values for MDR epidemics and use this to consider threshold criteria for eradication, the model should incorporate the emergence as well as transmission of resistance.

	Blower	**Blower (DR)**	**Vynnycky**	**Dye (DOTS)**	**Murray**	**Dye (AB use)**	**Dye (MDR)**	**Bahcall**
Reference	(Blower, McLean et al. 1995; Blower, Porco et al. 1998)	(Blower, Small et al. 1996; Blower and Gerberding 1998)	(Vynnycky and Fine 1997; Vynnycky and Fine 2000)	(Dye, Garnett et al. 1998)	(Murray and Salomon 1998)	(Dye and Espinal 2001)	(Dye and Williams 2000)	
Population size	Constant growth rate	Constant growth rate	Population growth matches actual population	Constant	Constant growth rate	Constant	Constant	Constant
DS	+	+	+	+	+	+	separately	+
DR	-	+	-	-	-	+	separately	+
MDR	-	-	-	-	-	+	+	+
Acquired resistance	-	Yes, but set as proportion treated	-	-	-	+	-	+
Primary resistance	-	+	-	-	-	+	+	+
R or R_0 defined	+	+	-	-	-	-	+	+
Immunity	-	-	+	+	+	-	-	-
Reinfection	-	-	+	?	+	?	+	+

Age structured	-	-	+	+	-	-	-	+
Control								
Treatment	+	+	-	+	+	+	+	+
Chemoprophylaxis	+	+	-	?	+	-	-	-
Case detection								
2nd line resistance treatment								
Parameter estimates from	Literature	Literature	Fit to data in England and Wales	Literature	Literature	Global surveillance data to estimate rates acquired resistance	Literature	Literature

Table 4-1. Summary of the seven models reviewed in this section and the model presented in this thesis, comparing critical model components.

VII. Conclusion

We have surveyed the literature on mathematical modelling of tuberculosis epidemics in this section in order to provide background into the basis of the model developed in the following sections. We have highlighted in Table 4-1 the key elements of models considering resistance within TB epidemics, and these elements will be explored further through the model development in Chapter 5 and sensitivity analysis in Chapter 6.

5. Chapter 5

Model for the control of resistant TB

5. CHAPTER 5

MODEL FOR THE CONTROL OF RESISTANT TB

I. GOALS

II. ABSTRACT

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IV. B. 1. ESTIMATES OF RESISTANT STRAIN TRANSMISSION
IV. B. 2. RELATIVE FITNESS RESISTANT STRAIN
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V. F. R FOR THREE STRAIN MODEL, NON-INTERACTING STRAINS, WITH REINFECTION
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VI. TREATMENT OUTCOMES: CRITICAL VALUES

VI. A. CRITICAL TREATMENT RATES FOR ERADICATION ALL TB STRAINS
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VII. AGE STRUCTURE

VII. A. AGE STRUCTURED MODEL
VII. B. SIMULATIONS
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VIII. MODEL PARAMETER VALUES

IX. SUMMARY OF FINDINGS

IX. A. MODEL FORMULATION
IX. B. MATHEMATICAL DEFINITIONS

I. Goals

I. A. To formulate a deterministic compartmental mathematical model for the acquisition of resistance and transmission of both sensitive and resistant TB strains.

I. B. To examine the dynamics of resistance within TB epidemics.

II. Abstract

A mathematical model for the emergence and transmission of resistant tuberculosis is developed in order to explore the dynamics of resistance within epidemics of TB. The compartmental deterministic model includes the emergence of acquired drug resistance during treatment as well as the transmission of primary resistance, within the context of a drug sensitive epidemic. The model includes three strains (one drug sensitive, one with single drug resistance, and one MDR), and allows for infection as well as reinfection. The basic and effective reproductive number for each strain in the absence (R_0) and presence of treatment (R), respectively, are defined. The critical treatment rates for strain eradication are derived, showing the balance between notification and cure rates.

Resistance is seen to emerge, in the model simulations, as series of linked sub-epidemics. The MDR epidemic emerges from a drug sensitive epidemic through acquired resistance, and these remain linked through treatment. After the initial emergence of acquired resistance during the early stages of a resistant epidemic, most of new resistant cases are due to primary transmission.

III.Methods

III. A. Simulations

Model simulations were run in Berkeley Madonna and C++. The set of ordinary differential equations for the model were resolved with Runge Kutta (RK4) method with a time step of 0.05 year. The size of the time steps was examined but did not significantly affect the numerical output. These differential equations, a flow chart of the model, and the parameter names and values are shown in Appendix 1.

III. B. Formulating the model: Epidemiological model for the spread of TB

Susceptible individuals are infected at rate equal to the force of infection (λ). The force of infection, λ, is equal to the proportion of infectious cases ($\frac{I}{N}$) multiplied by the effective contact rate (c). The effective contact rate is equal to the probability of contact multiplied by the probability of transmission.

Equation 5-1

$$\lambda = c\frac{I}{N}$$

A proportion p of infected individuals develops primary TB, defined as progression to active disease within five years of infection. The remaining infected individuals (1-p) move into a latent class (L) and then progress to disease at rate v.

A fraction i of disease progressions lead to an infectious case, with the remaining (1-i) resulting in non-infectious (NI) TB. Non-infectious TB cases have fewer symptoms, do not experience an increased mortality, and do not receive treatment. NI cases self-cure at a rate κ, the same as for infectious cases.

Infectious cases of TB (I) are detected at a notification rate (n) and are treated with a success rate of Φ. Infectious cases also self-cure at a rate κ, while treated and cured cases relapse to disease at a rate w.

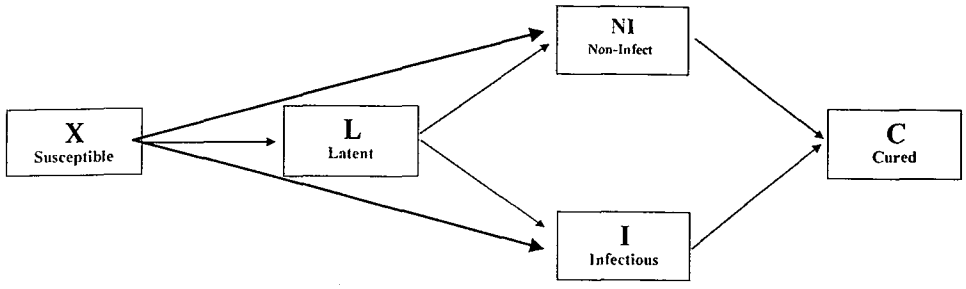


Figure 5-1. *Model for the transmission of a single strain of tuberculosis.* Infectious cases (I) transmit infection to susceptible individuals (X) in the population. Newly infected cases either progress directly to active disease or remain latently infected (L) and experience a slow rate of reactivation to disease. New cases of active disease are either infectious (I) or non-infectious (NI), and both may self-cure and become 'cured' (C). Infectious cases also receive treatment which results in faster cure. Recovered individuals (C) may relapse to disease.

III. C. Model for primary transmission of resistance

The model is expanded to include three strains, accounting for drug sensitivity (S), resistance to one drug (R), and resistance to two or more drugs (MDR). Susceptible individuals can be infected with a sensitive (S), singly resistant (R), or multiply resistant (M) TB strain at a rate of λ_S, λ_R, or λ_M accordingly. The strain-specific force of infection is defined as given for drug sensitive strains (λ_S) in Equation 5-2. The force of infection varies by strain depending on both the strain-specific contact rate and the prevalence of infectious cases.

Equation 5-2

$$\lambda_S = c_S \frac{I_S}{N}$$

Infectious cases of TB (I) are sensitive (I_S), singly resistant (I_R), or multiply resistant (I_M), and are detected at a notification rate (n). Sensitive cases are treated with

a success rate of Φ_S. Singly resistant cases are treated with a lower efficacy of Φ_R, and multiply resistant cases at an even lower efficacy of Φ_M. Infectious cases also self-cure at a rate κ, while treated and cured cases relapse to disease at a rate w. Non-infectious cases are considered to be strain-independent, as these cases do not receive treatment or transmit disease.

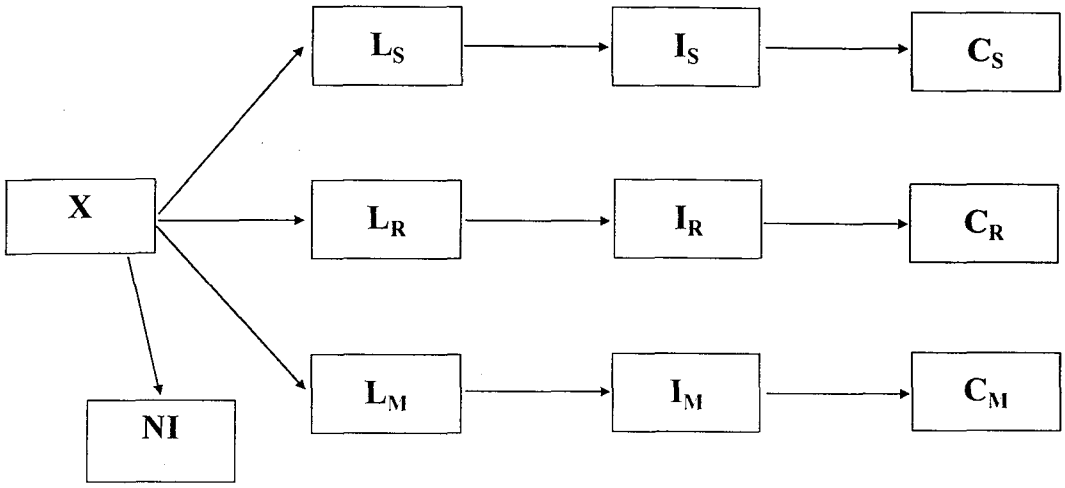


Figure 5-2. Model expanded to include the transmission of three strains, including a drug sensitive (S), a singly resistant (R), and a multiply resistant (M) strain. Each strain follows the transmission dynamics as outlined in Figure 5-1. Susceptible individuals (X) are subject to infection by an S, R, or M strain. Infected individuals either progress directly to active disease or remain latently infected (L) and are subject to a slow rate of reactivation disease. Active cases may be either infectious (I) or non-infectious (NI). Note this diagram does not reflect the reinfection included in the model.

III. D. Acquired resistance: treatment and emergence of resistance

There are three outcomes for the treatment of a drug-sensitive case (I_S): cure, failure with drug sensitivity, and failure with drug resistance. Treated I_S cases are successfully cured at a rate Φ_S. A proportion r of the $(1-\Phi_S)$ cases that fail treatment acquire single resistance. The remaining $(1-r)(1-\Phi_S)$ fail treatment and remain drug sensitive. This last class includes individuals with chronic drug sensitive TB, who remain sputum smear positive after repeated treatments.

Similarly, treated singly drug resistant cases (I_R) are cured at a rate Φ_R. A proportion r_2 of treatment failures gain a second resistance mutation and move into the I_M class, while the remaining treatment failures remain in I_R.

A treated MDR case has only two of these outcomes—cure (at rate Φ_M) or treatment failure, remaining in the I_M class. The probability of resistance emerging given treatment failure (r) depends upon the type of treatment and control program in place.

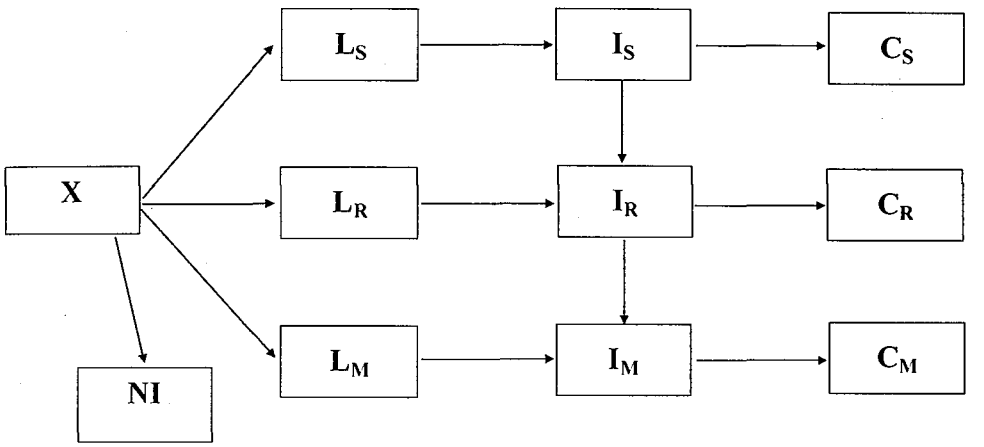


Figure 5-3. *Model expanded to include the emergence of acquired resistance during treatment.* Treated infectious drug sensitive cases (I_S) are either cured (C_S), fail treatment and remain drug sensitive (I_S), or fail treatment and acquire resistance (I_R). Similarly, treated singly resistant cases (I_R) are either cured, fail treatment and remain singly resistant (I_R), or fail treatment and acquire higher levels of resistance (I_M).

III. E. Model for reinfection

In the model, latently infected or previously treated individuals are subject to reinfection. Previously infected individuals are subject to the same force of infection as are non-infecteds, but may gain partial protection ($1-\gamma$) from developing exogenous disease. This protection is considered to be independent of strain type, such that latently infected individuals are equally susceptible to reinfection with any strain. For simplicity, only those reinfection events which successfully lead to primary progression

and exogenous disease are considered. Thus, previously infected individuals experience exogenous disease at a rate of $\gamma\, p\, (\lambda_S + \lambda_R + \lambda_M)$.

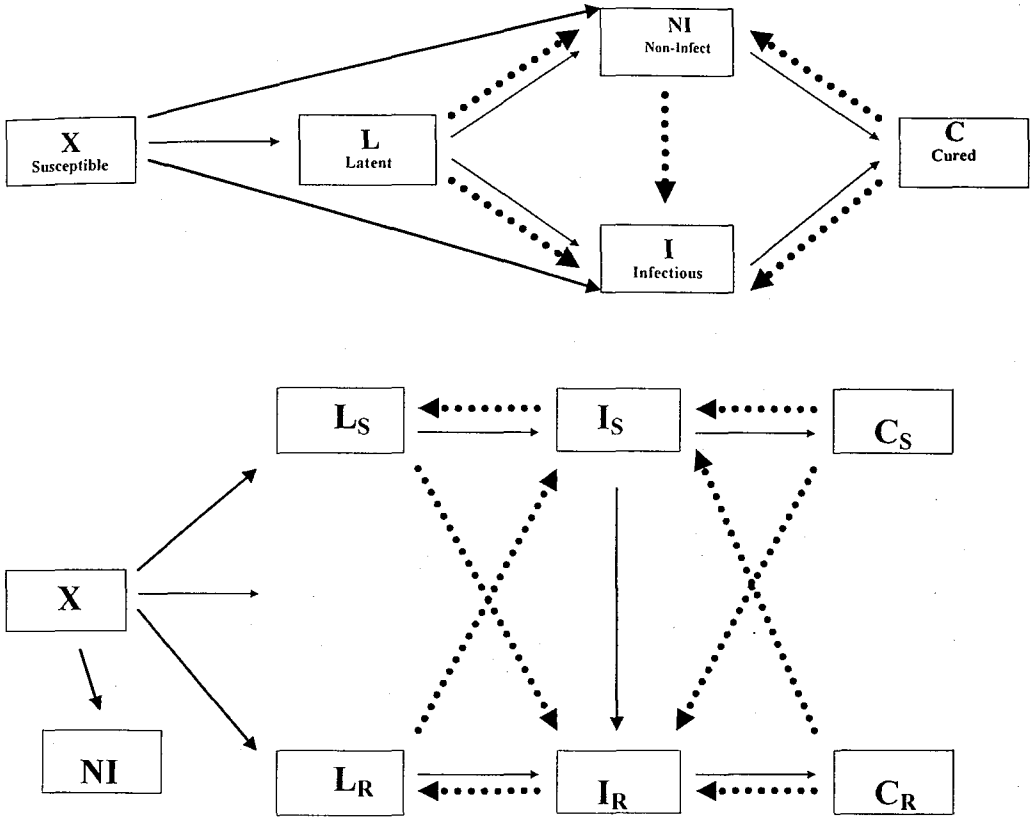


Figure 5-4. ***Model including reinfection.*** The model is shown to include reinfection for (A) a single strain model and (B) a two strain model. The dashed lines reflect routes of reinfection.

IV. Model Parameters

This section reviews the basis for the key model parameter estimates. As noted, these are based on reviews of literature and of previous models, noting their parameter estimates, as some new analyses. These parameter estimates are summarized in **Table 5-5**, which also notes the sources.

IV. A. Basic disease parameters

(1) Progression to disease

Only a small percentage of individuals infected with TB ever develop disease. A proportion of these progress rapidly to primary disease, defined as disease occurring within five years after initial infection. Estimates for the proportion developing primary disease range from 5-15%, and this is the basis for the parameter range used in the current model estimates (**Table 5-5**). The rates of progression to primary disease are higher in immune compromised individuals, in particular due to HIV coinfection, as well as in the elderly. (Davies 1998)(Bass 1990 ARRD)(Enarson and Roullin 1998)

Estimates for the risk of progression to disease can be found from epidemiological studies. The annual risk of infection was estimated from records of tuberculin sensitivity of adult male military recruits in the Netherlands between the 1930s and 1969. Using these estimates of the annual risk of infection, and notification data for the Netherlands, the risks of developing primary, reactivation, and exogenous disease were then estimated. These studies showed the highest risk of developing primary disease in the first year after infection, declining slightly over five years after infection, and then stabilizing at a low rate. It was assumed that there was a characteristic risk of progression to primary disease within the first five years after initial infection, distinct from progression to endogenous reactivation disease after five years. The annual risk of developing primary disease for the first five years after initial infection was estimated at 5.06% for adult males. After the first five years after primary infection, the risk of developing disease decreased significantly. The average annual rate of progression to endogenous reactivation disease was estimated at 0.0253%. (Sutherland, Svandova et al. 1982; Sutherland, Bleiker et al. 1983)

Subsequent studies estimated the risks of disease progression by fitting this data set to their own models. One study estimated the proportion of incident cases progressing to primary disease at 14% (range 8-15%), slightly higher than original estimates. (Dye, Garnett et al. 1998) In another study, the risks of progression to disease were estimated from data in England and Wales. The risk of primary progression after first infection was estimated at 8.6% in the first year, with a 13.8% cumulative risk in the first five years. The annual risk of reactivation to endogenous disease was estimated at 0.03%. (Vynnycky and Fine 1997)

In the model develop here, I use estimate for the proportion progressing to active disease of 5-15%, based on the previous model estimates reviewed above. For the rate of endogenous reactivation use 0.03%, similar to the Sutherland et al. estimates.

(2) Effective contact rate (ECR)

The effective contact rate (c), as defined in the model, is the number of people effectively contacted by each case. The commonly used estimate is such that each case infects on average 10 individuals per year. This estimate comes from studies by Styblo on TB incidence in the Netherlands. The annual risk of infection was correlated to the incidence of smear positive cases. Assuming an average case duration of infectiousness of two years, each smear positive case would contact and infect on average 10 individuals per year. (Styblo, 1991) More recent models have used this estimate for the ECR, with values ranging from 5 – 15 contacts per year. (Blower, McLean et al. 1995), (Porco and Blower 1998), (Dye and Williams 2000; Dye and Espinal 2001) Similarly to these previous model estimates, I use parameter range for the effective contact rate of 5-15 cases / year (average of 10).

(3) Case fatality and self-cure rates

The case fatality rate for active TB cases has been significantly reduced over the past century. This is related both to improvements in general health care and to the advent of chemotherapeutic treatment. In the prechemotherapy era, the average case duration of infectiousness was estimated at two years. Estimates of the mortality and cure rates, in the absence of treatment, are found in several sources. One source reports that in the absence of treatment, 50% of cases die within five years, 30% are cured, and 20% remain chronically infectious. (Grzybowski 1991) From another source, in the absence of treatment 50% of cases die within five years, 25% are cured, and 25% remain infectious. (Davies 1998) The results of early studies reporting the clinical course of patients in sanatorium, in the pre-chemotherapy era, can be found summarized in Table 5-1, reproduced from (Enarson and Hopewell 1998). This was the most useful source for estimating mortality and cure rates, as it reports the proportion of cases dead or recovered at 18, 36, and 60 months since diagnosis of disease. The model was fit to the data set shown in Table 5-1 for the proportion of cases which self-cure, die, or remain infectious over the course of five years after onset. The model fit is given in Equation 5-3, where I represents smear positive cases, and the parameters for the mortality rate (μ), the TB mortality rate (μ_T) and the self-cure rate (κ). As only a proportion of cases were shown in this data set, it was assumed a population of size 100 in order to fit model to data. This allowed fitting of the maximum likelihood estimate (MLE) and calculation of confidence intervals, although it should be noted that the confidence interval depends upon the actual population size of the data set. The parameters for the TB mortality rate (μ_T) and the self-cure rate (κ) were fit, with maximum likelihood estimates (MLE) of $\mu_T = 20\%$ and $\kappa = 17\%$. (Figure 5-5)

Equation 5-3

$$\frac{d(I)}{dt} = -(\mu + \mu_T + \kappa)\, I$$

Time since diagnosis (months)	Remain smear positive	Dead	Recovered
18	42%	30%	28%
36	25%	40%	35%
60	18%	49%	33%

Table 5-1. ***The clinical course of untreated tuberculosis.*** With increasing time since diagnosis, the proportion of cases remaining smear positive declined, while the proportion dead or recovered increased. At five years since diagnosis, 49% of cases have died, and 33% have recovered. Source: (Enarson and Hopewell 1998)

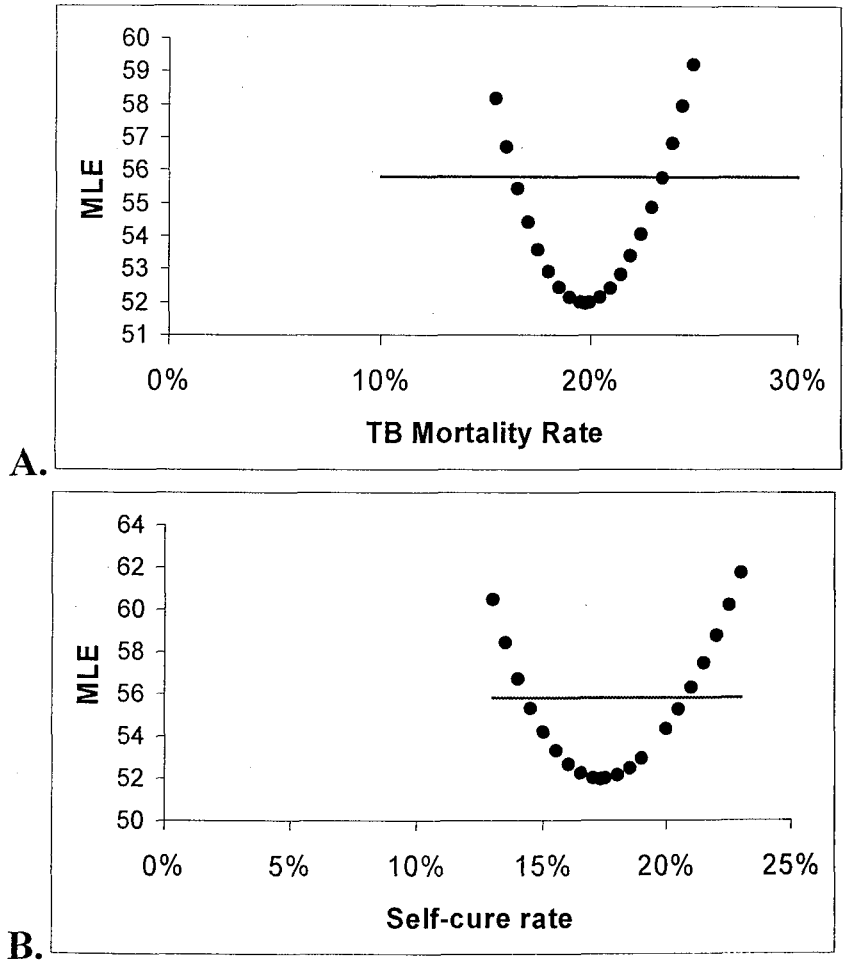


Figure 5-5. *Parameter estimates for the TB mortality and self-cure parameters.* The model was fit to the data set shown in **Table 5-1** for the proportion of cases which self-cure, die, or

remain infectious over the course of five years after onset. The MLE estimates for the TB mortality rate (μ_T) and the self-cure rate (κ) were μ_T = 20% and κ = 17%.

A. Estimate for the TB mortality rate (μ_T). The maximum likelihood estimate (MLE) for the TB mortality rate (μ_T), from fitting the model to the data set in **Table 5-1**, is μ_T=20%. The 95% confidence interval for the MLE fit for the TB mortality rate is μ_T = 16-23% (solid line).

B. Estimate for the self-cure rate (κ). The maximum likelihood estimate (MLE) for the TB self-cure rate (κ), from fitting the model to the data set in Table 5-1, is κ = 17%. The 95% confidence interval for the MLE fit for the self-cure rate is κ = 13-20% (solid line).

(4) Infectious disease

Infected individuals who progress to active disease may develop either infectious or non-infectious disease. Infectious disease occurs in the lungs in aerated caseous foci such that causes cases emit infectious particles in sputum. Non-infectious cases do not emit infectious particles in sputum and are therefore unable to transmit infection. The treatment regimens recommended for smear negative patients tend to be of shorter duration and require fewer drugs. Smear negative cases also experience reduced mortality. The proportion developing infectious disease has been difficult to estimate because of limitations both in case reporting and diagnosis. First, there may be differential reporting of infectious and non-infectious cases. Non-infectious cases tend to have fewer symptoms and cases may not seek medical treatment. In addition, non-infectious cases are more difficult to diagnose. Secondly, infectiousness is clinically classified based on a positive sputum smear or culture test. Due to limitations of these tests in detecting infectious particles in the sputum, the reverse does not hold, and a proportion of smear negative patients have been shown to transmit infection. (Davies 1998; Fishman and Elias 1998)

IV. B. Primary Resistance

IV. B. 1. Estimates of resistant strain transmission

The relative transmission rate of resistant, and in particular MDR, strains remains a source of significant uncertainty. Experiments have suggested significant variation in the transmission rates of resistant strains, ranging from reduced to greater than drug sensitive strains. In one study, the contacts of source cases with either sensitive or resistant TB were examined, and similar risks of infection and disease were found amongst contacts of either type. Higher risks of infection were found only amongst resistant cases which were previously treated. (Snider, Kelly et al. 1985) A recent study in Brazil also found similar risks of infection and disease for drug sensitive and resistant source cases, with about 40% of household contacts infected and 4% with active disease. (Teixeira, Perkins et al. 2001) Another study used contact tracing combined with RFLP analysis to show MDR transmitted to contacts at similar rates. In another study, 6% of contacts with MDR patients were found to convert to active disease. (Nitta, Knowles et al. 2002) This can be compared to another study in which 4% of national and 8% of foreign born contacts of drug sensitive cases converted. (Marks, Taylor et al. 2000) These studies together suggest similar rates of conversion amongst contacts of sensitive and resistant source cases.

Studies have also investigated the proportion of resistant cases due to ongoing transmission, by two different methods. The first method involves estimating the prevalence of resistance amongst new and previously treated cases, assuming that new cases are caused by primary resistance, and previously treated strains by resistance acquired during treatment. This classification was used in initial studies of resistance. (WHO/IUATLD 1997) Later studies, using RFLP analysis and contact tracing, revealed complications in making this distinction. The proportion of acquired resistant

cases were being overestimated, as some of the previously treated cases appeared to have been initially infected with resistant cases. (Van Rie, Warren et al. 2000) In order to correct for this, later surveillance reports referred to the proportion of resistant cases amongst new and previously treated cases, without inferring from this the cause of resistance. (WHO/IUATLD 2000) In general, contact tracing or strain typing are required to differentiate acquired and primary resistance.

More recently, convention has led towards using clustering of strains as a surrogate for recent transmission, and such analysis has been used to estimate the relative proportion of resistant strains in a population due to ongoing transmission. In an area of high incidence (Capetown, South Africa), the proportion of resistance due to primary transmission, as detected by RFLP strain typing, was high; over 50% of MDR cases were clustered. (Van Rie, Warren et al. 2000) In Denmark, over 70% of resistant strains were found to be clustered. (Thomsen, Bauer et al. 2000) There have also been reports of 'super fit' resistant strains which have been found more frequently in clusters and are believed to have greater transmission. (Agerton, Valway et al. 1999) (Toungoussova, Sandven et al. 2002) (Kruuner, Hoffner et al. 2001)

Other strain typing studies have found resistant cases less likely to be found in clusters. (Samper, Iglesias et al. 1998; Garcia-Garcia, Jimenez-Corona et al. 2000) (Nitta, Knowles et al. 2002) Cases with a particular INH resistance mutation were found in clusters less frequently than drug sensitive strains. (Van Soolingen, Borgdorff et al. 1999)

IV. B. 2. Relative fitness resistant strain

In order to account for possibly different transmission rates, each strain in the model is given a distinct transmission parameter. It is assumed that single drug resistance does not confer a significant fitness cost, but that MDR may have reduced

transmissibility ($c_S = c_R > c_M$). Further consideration of the fitness of resistant strains and the implications of relative fitness on within host dynamics under chemotherapy treatment is explored within a model and simulations found in Appendix 5.

IV. C. Acquired resistance rates

In the model, cases failing treatment may either remain chronically infectious or gain resistance. The probability of acquiring resistance, given treatment failure, is equal to the parameter *r* (the acquired resistance rate).

(1) Clinical evidence for resistance amongst treatment failures

Previous treatment has been shown repeatedly to be a primary risk factor for resistance. (Borchardt, Kirsten et al. 1996; Janmeja and Raj 1998) (Iseman 1999) As shown in Table 5-2 the proportion of cases with acquired resistance increases with the number of previous courses of treatment. In this trial, of cases having received one course of treatment, 33% had resistance to one drug and 22% MDR. Amongst cases which received two courses of treatment, 64% had developed resistance to one drug and 55% MDR. Amongst cases which received three courses, 100% had single resistance and 97% MDR. (Chaulet and Zidouni 1998)

Courses of treatment	Patients assessed	Resistant to at least one drug		Resistant to INH and RIF at least (MDR)	
One	27	9	33%	6	22%
Two	22	14	64%	12	55%
Three	32	32	100%	31	97%
Total	81	55	68%	49	60%

Table 5-2. *Acquired bacterial resistance in 81 patients previously treated by one or more courses of chemotherapy containing isoniazid and rifampicin for 6 months;*

Algeria 1992. Source: (Mazouni, Zidouni et al. 1992) cited in (Chaulet and Zidouni 1998)

(2) Estimates from surveillance and model

Estimates found in the literature suggest that acquired resistance accounts for only a small proportion of treatment failures, and that cases are more likely to fail treatment for other reasons. For example, in one study, the proportion of treatment failures acquiring resistance was estimated from surveys on the resistance prevalence amongst new and previously treated cases. The regression line for the incidence of resistant to sensitive cases amongst previously treated cases was taken, with the slope assumed to equal the relative treatment cure rate for resistant cases, and the intercept the proportion which acquired resistance. This estimated resistance amongst treatment failures at a rate of 13% for isoniazid, 8% for rifampicin, and 7% for MDR. This study suggests that rates of acquiring single resistance may be low because many singly resistant cases are cured by SCC, and are more likely to fail treatment for reasons other than resistance. (Dye and Espinal 2001)

In order to estimate the appropriate rates of acquired resistance to use in our model, simulations were run to compare predicted and observed resistance prevalence. The notification, treatment, and cure rates in several different locations were used to simulate a model epidemic. The rate of acquired resistance (r) was varied such that the observed prevalence of resistance was matched by the model. All other intrinsic model parameters (those not determining part of the control strategy) were held constant. The effect of jointly varying other parameters is examined in Chapter 6. Model simulations were performed to simulate the scenarios represented in Table 5-3. Within each location scenario, the model parameters for control program were also held constant. In each example, the cure rates for sensitive and resistant cases were used as shown. The notification and treatment rates were used as given in recent surveillance reports

for that region. (WHO 2003) In order to reproduce close to the given prevalence of single resistance and MDR, an acquired resistance rate (r) of 5-15% was used in the model.

In Peru and Hong Kong, there was 100% reporting under DOTS and strong first line control including high treatment and cure rates. In both of these scenarios, it was difficult to fit the resistance prevalence observed to the given treatment and cure rates. This most likely reflects the recent improvements in program control in these regions, which followed years of much poorer treatment practices, that had resulted in the buildup of resistance levels. Under the strikingly poor cure rates found in Ivano Oblast, a higher rate of acquired resistance was used in order to match the higher resistance prevalence found. This is likely to reflect higher probability of acquiring resistance under poor control practices.

A noted limitation of these simulations is the assumption that the control program parameters are constant over time. While this will matter less in high incidence regions where most of incidence due to recent transmission, this can have a stronger effect in those low incidence regions in which control program improvements have sharply reduced incidence. In such cases, much of new incidence may be due to reactivation of latent infection acquired years to decades earlier, under poorer control programs/ conditions. As the regions noted in Table 5-3 do not fit such criteria (they remain largely high incidence regions) this may hold for these regions. However, in Peru and Hong Kong, this may account for different estimates of R.

	DS cure rate	DR cure rate	MDR cure rate	Prevalence single resistance	Prevalence MDR
Location	**Source: [1]**			**Source: [2]**	
Korea	85%	75%		10.6%	2.2%

Peru	85%	79%	58%	18%	3%
Hong Kong	89%	80%	60%	12.2%	1.4%
Ivano Oblast	37%	24%		32.4%	9%
Dominican Republic	28%	23%		------	------
Italy	63%	43%		12.3%	1.2%

Table 5-3. *The cure rates and prevalence of resistance from surveillance in 6 locations.*

[1] Shown is the cure rate for drug sensitive, singly resistant, and MDR cases amongst all (new and previously treated) smear positive cases. Source for the cure rates in these 6 countries. (Espinal, Kim et al. 2000)

[2] Shown is the prevalence of single resistance and MDR amongst all cases. No data was available for the Dominican Republic, as this country was not reporting to the WHO under DOTS. Source for the prevalence of resistance in these 6 countries: (WHO/IUATLD 2000).

(3) Control program and quality of treatment

These estimates of the proportion of acquired resistance have been made assuming that there is an inherent risk of acquiring resistance during treatment. However, there is also evidence that this statistic depends upon the quality of treatment. Under good control measures, there is a low rate of acquired resistance. When the control is poor, higher levels of resistance are found, particularly when there are lower rates of DOTS implementation. (WHO/IUATLD 2000)

The rates of acquired resistance are correlated to the quality of treatment, including regular drug supply and supervision of therapy. Higher rates of resistance have been associated with errors in treatment and case management. (Mahmoudi and Iseman 1993) Poor treatment practices correlated to increased resistance rates include adding one drug to a failing regimen, using an inadequate regimen, not diagnosing initial resistance, and poor patient adherence. (Iseman 1999) Use of supervised DOT

treatment combined with regulated drug supply has been shown to reduce the proportion of cases developing resistance. (Weis, Slocum et al. 1994)

IV. D. Reinfection

Individuals previously infected with tuberculosis are subject to reinfection. The possibility of reinfection has been long recognized. (Romeyn 1970; Canetti, Sutherland et al. 1972). Patients have been observed to relapse with disease of a different drug resistance pattern. Most notably, patients treated for drug resistant TB have been seen to relapse with a drug sensitive case. Before molecular typing experiments were common, it was difficult to distinguish whether these cases were due to reversion of the drug resistance mutation or reinfection with a drug sensitive strain. (Stewart 1966) There have also been examples of patients with drug sensitive disease developing drug resistance as the result of exogenous reinfection with a resistant strain. (Small, Shafer et al. 1993)

More recent experiments have provided further evidence for reinfection of tuberculosis patients. Molecular typing techniques have been used to compare strains collected from the same patient during initial and subsequent occurrences of disease. (Fine and Small 1999; Van Rie, Warren et al. 1999; de Boer AS 2000) When the RFLP pattern has significantly changed between collections, it is assumed that disease has been caused by a new strain. This is a reasonable assumption because the timescale for RFLP change is very long, with the pattern remaining constant between cycles of transmission. In particular, there seems to be a high degree of stability of IS6610 typing. (Niemann, Richter et al. 1999; Van Soolingen 2001) Using such methods, numerous studies have found examples of reinfection occurring in a wide range of both low and high incidence regions.

In high incidence regions, individuals are subject to a high annual risk of infection, and may be reinfected multiple times. However, current detection methods cannot differentiate the number of times an individual has been infected, or reveal which infection (or combination of infections) may have triggered disease progression.

While reinfection may not greatly impact the overall prevalence of disease in low incidence countries, there may be a significant effect in localized areas with individuals at higher risk for reinfection. In particular, immigrants have been found to have a greater risk of reinfection, as do patients with HIV coinfection. For example, immigrants from high incidence countries may live in the same community, effectively creating a high incidence region. (Bandera, Gori et al. 2001) (Murray and Nardell 2002)

Previous models have also suggested the importance of reinfection in understanding the transmission dynamics of tuberculosis. Sutherland estimated the risk for exogenous disease at 1.91% annually for five years following reinfection. (Sutherland, Svandova et al. 1982) Vynnycky has shown that models which include partial susceptibility to reinfection provide a significantly better fit to observed trends in the decline of tuberculosis cases (within the older age classes) over the past century in the low incidence areas of England and Wales. Previous infection was estimated to confer approximately 40% protection from developing exogenous disease. (Vynnycky and Fine 1997; Vynnycky and Fine 2000).

V. Mathematical definitions for model epidemic

V. A. Defining the basic and effective reproductive number

The basic reproductive number, R_0, is defined as the average number of new cases caused by introducing one infectious case into an entirely susceptible population. This is a measure of reproductive potential as well as fitness. However, this measure does not hold in the later stages of an epidemic, when the population contains infected, infectious and cured individuals, each with differing levels of susceptibility to infection and disease. In such situations where the initial population is not entirely susceptible, a related measure, termed the effective reproductive number (R), is used to describe reproductive potential. R can also be defined within populations where the susceptible class is reduced through vaccination. . (Anderson 1991) Similarly, treatment can reduce the spread of infection and disease within the population. We therefore use the effective reproductive number (R) to describe the average number of new cases caused by one infectious case introduced into a susceptible population, in the presence of a control strategy. In this section, both the basic (under no treatment) and effective reproductive (under treatment) numbers are defined for the model described in the previous section.

For tuberculosis epidemics, the basic reproductive number (R_0) is equal to the product of the probability of an infection progressing to active disease (b), the effective contact rate (c) and the duration of infectiousness (τ). The effective reproductive number (R) has the same components; however, treatment may reduce the case duration of infectiousness (τ).

Equation 5-4

$$R_0 = b\, c\, \tau$$

For individuals infected with tuberculosis, there are two routes of progression to active disease: primary progression soon after infection, or latent infection followed by reactivation disease. The probability of a given infection leading to active disease (b) incorporates both possibilities.

The effective contact rate (c) provides a measure for the per capita rate of new infections caused per infectious case. The ECR is equal to the probability of a contact between an infectious and susceptible times the probability of transmission per contact. While the contact rate depends on host and community factors, the transmission rate is considered to be strain dependent. Thus, a distinct ECR is defined for each strain, assuming an equal contact rate but varying transmissibility.

The duration of infectiousness (τ) is a measure of the average time cases spend in the infectious state. The case duration of infectiousness incorporates all routes for a case to become non-infectious, including cure by either natural self-cure or through treatment, and death from tuberculosis or other causes. Prior to use of chemotherapy for treatment of tuberculosis, the average case remained infectious for two years. The use of effective drug regimens results in faster cure and thus reduces the case duration of infectiousness.

Solving the system of model equations at equilibrium allows for a measure of system dynamics. In addition, this allows a derivation of the basic reproductive number, which is inversely proportional to the proportion susceptible at equilibrium. In the examples given below, the model is solved for the proportion susceptible at equilibrium ($\overline{X}$). This is then used to derive the basic reproductive number (R_0) in the absence of treatment or the effective reproductive number (R) in the presence of treatment. These formulas are derived in scenarios with increasing levels of complexity. First, R_0 is derived for a model for the transmission of one or three strains of TB in the absence of treatment. Second, R is derived for a model for transmission of

one or three strains in the presence of treatment. Third, R is derived for a model which includes reinfection in the presence of treatment. Fourth, R is derived for a model which includes both reinfection and interacting strains.

Equation 5-5

$$\overline{X} = \frac{1}{R_0}$$

V. B. R_0 defined: in absence of treatment

In this first example, a model which includes the transmission of a single strain of TB, in the absence of treatment, was examined. The system of equations describing this model was solved at equilibrium for the proportion susceptible, and from this R_0 was derived.

Equation 5-6

$$R_0 = c\left(\frac{1}{\mu + \mu_T + \kappa}\right)\left(i \quad p + \frac{i\,(1-p)\,v}{\mu + \upsilon}\right)$$

The components of the reproductive number, as delineated above, are easily discernable in this equation. The probability of contact and transmission is given by the effective contact rate (c). The case duration of infectiousness is given by the inverse of the rate of mortality from natural causes (μ) or TB (μ_T) and the self-cure rate (κ).

Equation 5-7

$$\tau = \frac{1}{\mu + \mu_T + \kappa}$$

The proportion of infected individuals progressing to active disease is given by the sum of the proportion progressing to primary disease (immediately) and the

proportion progressing from latent to reactivation disease (at a rate v), all multiplied by the probability of developing infectious disease (i).

Equation 5-8

$$\boldsymbol{b} = i\ p + \frac{i\ (1-p)\ v}{\mu\ +\upsilon}$$

Next, a three-strain model is examined, and the R_0 values for each strain defined. In the absence of treatment, the three strains (sensitive, resistant, and MDR) remain unlinked, as treatment is required for acquired resistance. Thus, each strain is transmitted within separate epidemics in the population. The R_0 of each strain differs only by the effective contact rate (c). The average case duration of infectiousness, in the absence of treatment, is the same for all strains. The proportion progressing to active disease is also strain-independent.

Competition between the strains is determined by the relative transmissibility. The strain with the greatest ECR will dominate the epidemic at equilibrium. The time taken to reach this equilibrium state will depend upon the relative prevalence of each strain at the start of the simulation. For example, if the model simulation is begun with only drug sensitive cases, no resistance will ever emerge. Only if the simulation is begun with resistant strains will transmission cycles of resistance continue.

Equation 5-9

A. $$R_{0S} = c_S \left(\frac{1}{\mu + \mu_T + \kappa}\right)\left(i\ \ p + \frac{i\ (1-p)\ v}{\mu\ +\upsilon}\right)$$

B. $$R_{0R} = c_R \left(\frac{1}{\mu + \mu_T + \kappa}\right)\left(i\ p + \frac{i\ (1-p)\ v}{\mu\ +\upsilon}\right)$$

C. $$R_{0M} = c_M \left(\frac{1}{\mu + \mu_T + \kappa}\right)\left(i\ p + \frac{i\ (1-p)\ v}{\mu\ +\upsilon}\right)$$

V. C. R for one strain model

The effective reproductive number (R) in the case of a one strain model, assuming no reinfection, is defined here. The system of equations was solved at equilibrium for the proportion susceptible, and from this the R value was derived as below.

Equation 5-10

$$R = c \left(\frac{1}{\mu + \mu_T + \kappa + n\phi - (\kappa + n\phi) \frac{w}{\mu + w}} \right) \left(i\ p + \frac{i\ (1-p)\ v}{\mu + \upsilon} \right)$$

The components of the reproductive number are easily recognized in this definition, and similar to those for R_0 defined above. The contact rate c and the probability of progression to active disease (b) have not been altered by including treatment in the model, and remain the same as for the definitions of R_0 above. The duration of infectiousness (τ) has been changed by including treatment. The case duration of infectiousness is equal to the probability of mortality, self-cure, treatment cure, or cure followed by relapse. In model parameters, the duration of infectiousness is given as below. In this description, it is easy to see how improvements to control through increased treatment (n) and cure rates (Φ) reduce the duration of infectiousness.

Equation 5-11

$$\tau = \frac{1}{\mu + \mu_T + \kappa + n\phi + (\kappa + n\phi) \frac{w}{\mu + w}}$$

V. D. R for one strain model with reinfection

The reproductive number is defined here for a one strain model which includes reinfection. As in the previous example, the system of equations was solved at equilibrium for the proportion susceptible, and from this the R value was derived. The model assumes that previous infection confers some given protection (at a rate of 1-γ) from reinfection, but that reinfected individuals have the same probability of progression given infection or reinfection. This is reflected in the derived R value; introducing reinfection affects the duration of infectiousness (τ), but leaves unchanged the contact rate (c) and probability of progressing to disease (b). In particular, the case duration of infectiousness now incorporates the possibility for cured individuals to be reinfected and develop exogenous disease. By this definition, reinfection would extend the duration of infectiousness of the individual, through subsequent cases of infection and disease. While this may not apply to a strict definition of the duration of infectiousness of a single case, this does appear in the R value as reinfected individuals will stay in the infectious class of the model for a longer period. As shown, this depends upon the probability of a previously infected individual becoming reinfected and developing exogenous disease, given as $\gamma\ i\ p\ \lambda$.

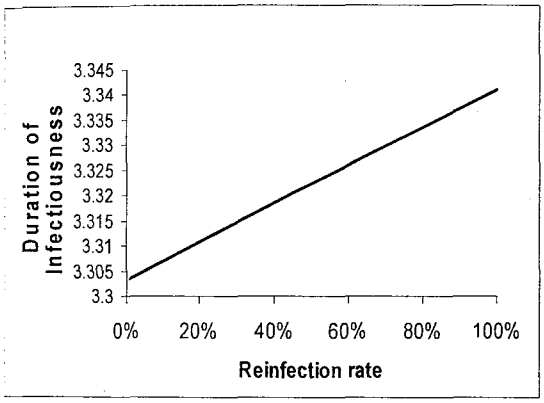
As explored in Figure 5-6, varying the reinfection parameter has a very small impact on the duration of infectiousness. Higher rates of reinfection increase the duration of infectiousness, but this is not significant when compared to the affect of treatment. The impact of reinfection on duration of infectiousness also depends on the prevalence of infectious disease in the population; the greater the prevalence of disease, the more significant the increase in DOI with the reinfection parameter. (Figure 5-6C) From this examination, it appears that varying the reinfection parameter may affect the

model epidemic when there is a high prevalence of disease and treatment rates, but has little effect under low prevalence. (Figure 5-6)

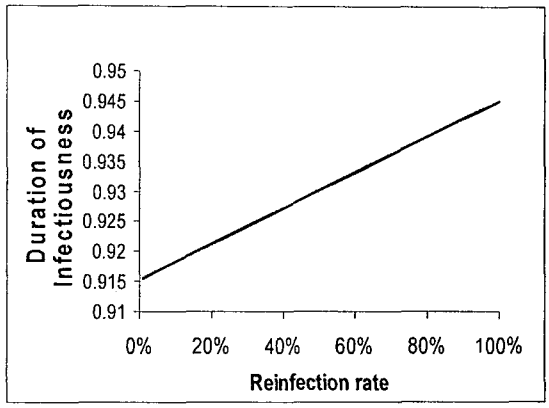
Equation 5-12

$$R = c \left(\frac{1}{\mu + \mu_T + \kappa + n\phi - (\kappa + n\phi)\frac{(w + \gamma i p \lambda)}{u + w + \gamma i p \lambda}} \right) \left(i p + \frac{i(1-p) v}{\mu + \upsilon} \right)$$

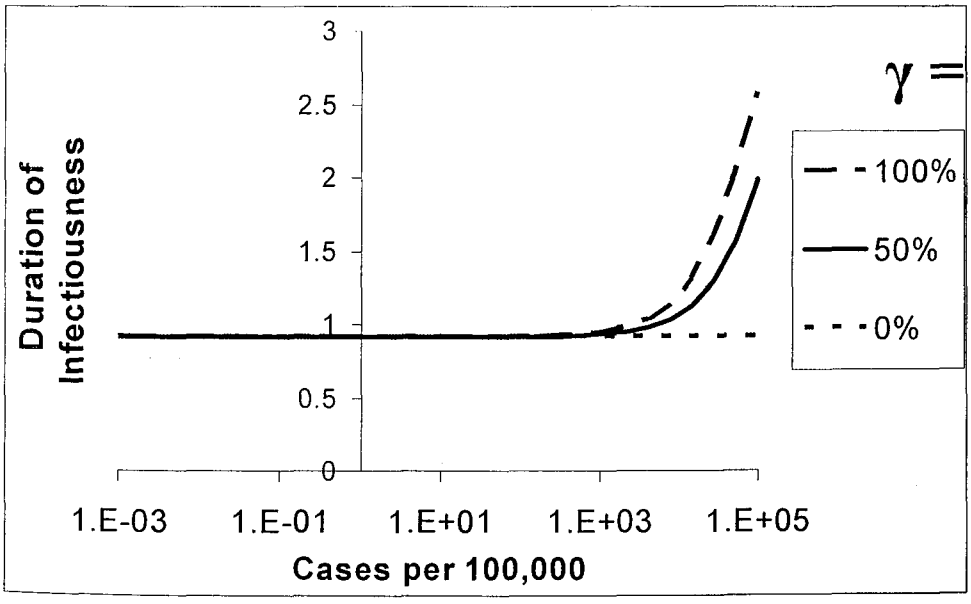
$$\tau = \frac{1}{\mu + \mu_T + \kappa + n\phi + (\kappa + n\phi)\frac{(w + \gamma i p \lambda)}{u + w + \gamma i p \lambda}}$$



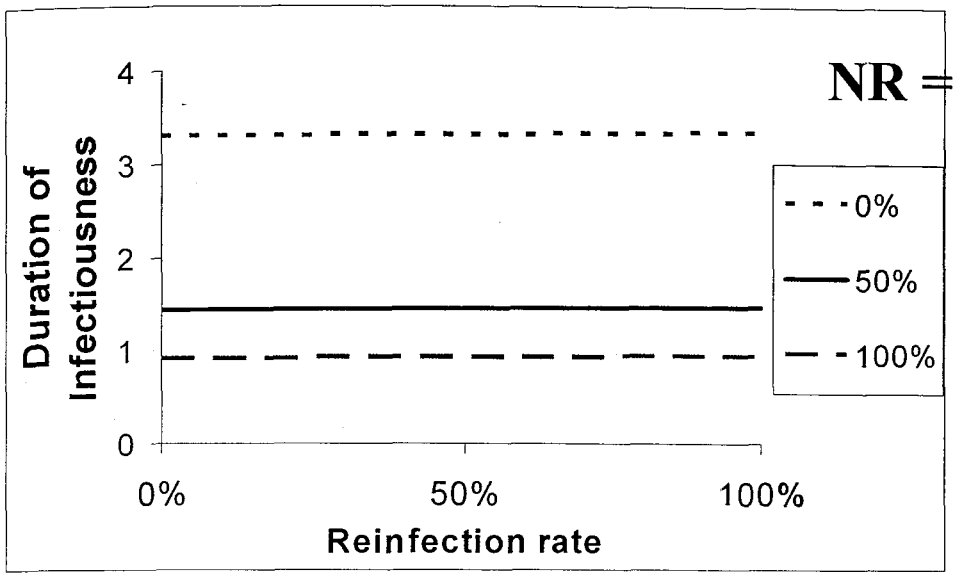
A



B



C



D

Figure 5-6. Reinfection parameter has small impact on duration of infectiousness, and depends upon the prevalence of infectious disease and the treatment rate.

A. No treatment. The notification rate was set at 0% and the prevalence of infectious cases at 1,000 cases per 100,000. Increasing the reinfection parameter results in a slight increase in the average case duration of infectiousness, from 3.305 years (at 0% reinfection) to 3.34 years (at 100% reinfection).

B. Treatment rate =100%. The notification rate was set at 100% and the prevalence of infectious cases at 1,000 cases per 100,000. Increasing the reinfection parameter results in a slight increase in the average case duration of infectiousness, from 0.915 years (at 0% reinfection) to 0.945 years (at 100% reinfection).

C. Prevalence of disease. When there is a higher prevalence of disease, there is an increased opportunity for reinfection, and increasing the reinfection parameter has a slightly greater influence on the duration of infectiousness. The prevalence of disease was varied from 10^{-3} to 10^{5} cases per 100,000. The notification rate was set at 100%. The susceptibility to reinfection was set at γ= 0%, 50%, or 100%, as shown in the legend.

D. Treatment rate has greater impact than reinfection on duration of infectiousness. The prevalence of infectious cases was set at 1,000 cases per 100,000. The notification rate was set at either NR = 0%, 50%, or 100%. The reinfection parameter was varied from 0% to 100%. Increasing the reinfection parameter had a small impact on the duration of infectiousness compared to increasing the notification rate.

V. E. R for three strain model, non-interacting strains, no reinfection

In this three-strain model, there are three unlinked sub-epidemics of TB, corresponding to the S, DR, and MDR strains. As each strain has distinct dynamics, a strain-specific reproductive number is defined.

Equation 5-13

A.

$$R_S = c_S \left(\frac{1}{\mu + \mu_T + \kappa + n\phi_S - (\kappa + n\phi_S)\frac{w}{\mu + w}} \right) \left(i\,p + \frac{i(1-p)\,v}{\mu + \upsilon} \right)$$

B.

$$R_R = c_R \left(\frac{1}{\mu + \mu_T + \kappa + n\phi_R - (\kappa + n\phi_R)\frac{w}{\mu + w}} \right) \left(i\,p + \frac{i(1-p)\,v}{\mu + \upsilon} \right)$$

C

$$R_M = c_M \left(\frac{1}{\mu + \mu_T + \kappa + n\phi_M - (\kappa + n\phi_M)\frac{w}{\mu + w}} \right) \left(i\,p + \frac{i(1-p)\,v}{\mu + \upsilon} \right)$$

The duration of infectiousness, shown here for drug sensitive cases, is inversely proportional to the death, self-cure, and treatment rates, as well as the proportion of cured cases which relapse to disease.

Equation 5-14

$$\tau_S = \frac{1}{\mu + \mu_T + \kappa + n\,\phi_S - (\kappa + n\,\phi_S)\frac{w}{\mu + w}}$$

Treatment reduces the case duration of infectiousness. Increasing the treatment rate reduces the duration of infectiousness for each strain depending upon the strain-specific cure rate. Treatment reduces the duration of infectiousness for sensitive cases, which experience higher cure rates, more than resistant cases. (Figure 5-7)

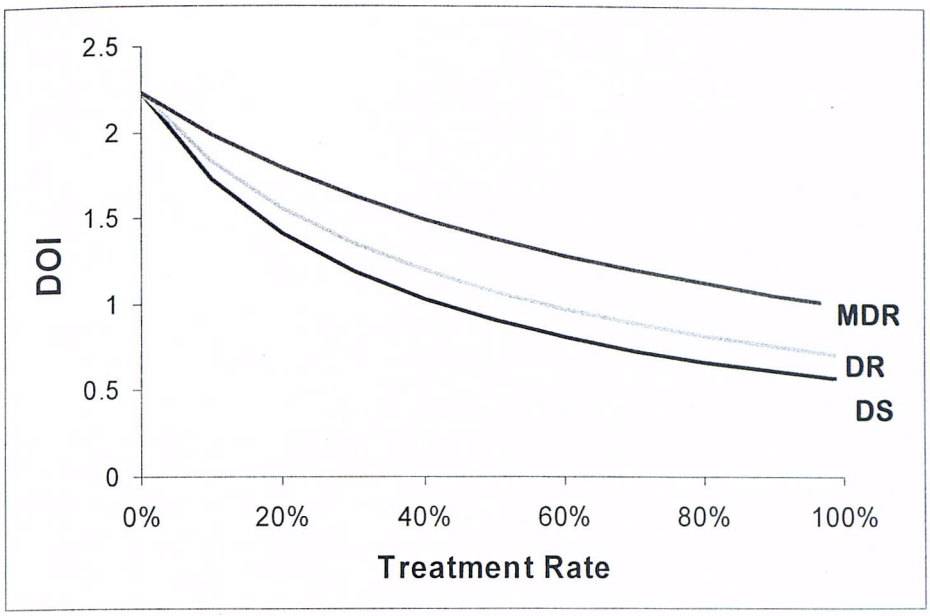


Figure 5-7. ***Treatment reduces the average case duration of infectiousness.*** Increasing the treatment rate (treatment rate, n) reduces the average case duration of infectiousness (DOI). When there is no treatment (n=0%), the average case duration of infectiousness is 2.2 years. As the treatment rate is increased, the DOI is reduced, depending upon the strain and treatment efficacy. The DOI of drug sensitive strains (DS) is reduced the most, followed by singly resistant (DR) and then MDR (MDR) strains, corresponding to the relative treatment efficacy. The treatment cure rate for DS cases is set at 90%, for DR cases at 70%, and for MDR cases at 40%.

The probability of an infection leading to a case of active, infectious disease is common for all strains. As defined above, the probability of progression (b) is equal to the probability of primary progression (p) and the probability of reactivation.

Equation 5-15

$$b = i\ p + \frac{i\ (1-p)\ v}{\mu\ + \upsilon}$$

This presentation makes it clear that the fitness difference between the strains is due to treatment (each strain has a different treatment efficacy Φ) and transmission (strain-dependent effective contact rate, c). Thus, in this model, the two variables which determine the relative fitness of the three strains include one inherent to the strain (c) and one to the control strategy (Φ). As discussed above, resistant strains may have a fitness cost manifest in a lower rate of transmission. This representation illustrates how changes in the transmission rate (c) directly affect the reproductive number and strain fitness.

V. F. R for three strain model, non-interacting strains, with reinfection

In this scenario, the model includes three unlinked strains, treatment, and the reinfection of previously infected individuals. This model does not include the emergence of acquired resistance during treatment. The solutions for the R values for each strain are shown below. As before, the difference in the R values between strains depends upon the contact rate and the duration of infectiousness. Also as in the previous example, increasing the notification rate and/or the strain-specific cure rate reduces the strain-specific duration of infectiousness. In addition, in this example, including reinfection will slightly affect the duration of infectiousness of each strain, as explored in the earlier example shown in Figure 5-6.

Equation 5-16

A.

$$R_S = c_S \left(\frac{1}{\mu + \mu_T + \kappa + n\phi_S - (\kappa + n\phi_S)\dfrac{(w + \gamma\ i\ p\ \lambda_S)}{u + w + \gamma\ i\ p\ \lambda_S}} \right) \left(i\,p + \frac{i(1-p)\,v}{\mu + \upsilon} \right)$$

B.

$$R_R = c_M \left(\frac{1}{\mu + \mu_T + \kappa + n\phi_R - (\kappa + n\phi_R)\dfrac{(w + \gamma\ i\ p\ \lambda_R)}{u + w + \gamma\ i\ p\ \lambda_R}} \right) \left(i\,p + \frac{i(1-p)\,v}{\mu + \upsilon} \right)$$

C.

$$R_M = c_M \left(\frac{1}{\mu + \mu_T + \kappa + n\phi_M - (\kappa + n\phi_M)\frac{(w + \gamma\ i\ p\ \lambda_M)}{u + w + \gamma\ i\ p\ \lambda_M}} \right) \left(i\,p + \frac{i(1-p)\,v}{\mu + \upsilon} \right.$$

V. G. R for three strain model, interacting strains

In this scenario, the interaction between the strains resulting from acquired resistance during treatment complicates the equilibrium solution. In place of a complete solution of R for each strain, approximations were made based upon the previous examples. While the complete model follows the trends as given in the previous example for unlinked strains, the emergence of acquired resistance affects the equilibrium dynamics.

Selection for resistance during treatment provides a link between the otherwise separate epidemics of each strain, shifting the equilibrium balance between the strains. When there are sensitive strains in the population which receive treatment, acquired resistance will increase the R value for the resistant strains. While the emergence of acquired resistance does not affect the transmission or duration of infectiousness of resistant strains, it does increase the base prevalence of resistance. Thus, at equilibrium, the same quantitative outcome, in terms of which strain will dominate the epidemic, will occur as given in the previous example. However, if at equilibrium sensitive strains remain and receive treatment, there will be a continuing low prevalence of resistance. New cases of acquired resistance will continue to emerge, and thus resistance cannot be eradicated unless either treatment is stopped or all infection is cleared from the population.

The inclusion of acquired resistance in this model also affects the point at which coexistence between the strains occurs. Coexistence is possible when the R values are equal and each greater than one. However, when acquired resistance increases the prevalence of resistance beyond its intrinsic fitness at equilibrium, then coexistence occurs when R_R is this small (and not defined here) quantity less than R_S.

VI. Treatment Outcomes: Critical Values

The effective reproductive number (R) can be used to predict the outcome of a control strategy in terms of the survival of each strain. There are three basic model outcomes: the eradication of all strains, the survival only of resistance, or the coexistence of both sensitive and resistant strains. It is impossible to remove resistance from the population as long as sensitive cases remain, because of the possibility of acquiring resistance during treatment.

The model outcomes are shown here in terms of strain R values. This is shown first for a model with one resistant strain (R, Table 5-4A and Figure 5-8), as a simpler example of the three possible outcomes. Next, the model outcomes are shown for a model with two resistant strains (R and M, Table 5-4B). In either scenario, the same three possible outcomes exist, as it is impossible to remove either single or multiple drug resistance from the population while drug sensitive cases remain. All TB in a population will be eradicated only if R of each strain is less than one. If R of any of the strains is greater than one, and the population receives treatment, resistance will continue.

Table 5-4

A.

R_S	R_R	Outcome

<1	<1	Eradicate all TB strains
<1	>1	Only R_R persists
>1	$R_S < R_R$	Coexistence

B.

R_S	R_R	R_M	Outcome
<1	<1	<1	Eradicate all TB strains
<1	>1	>1	Only resistance persists
		$R_R < R_M$	MDR dominates
	$R_M < R_R$		DR dominates
>1	$R_S < R_R$	$R_S < R_M$	Coexistence sensitive and resistant
		$R_R < R_M$	MDR dominates
	$R_M < R_R$		DR dominates

Table 5-4. *The model equilibrium outcome based on the absolute and relative values of the effective reproductive number (R) for each strain.* Three outcomes are possible under any given treatment strategy: the eradication of all strains, persistence of only drug resistance, or coexistence of drug sensitive and resistant strains. These three qualitative outcomes are the same whether a two (A) or three (B) strain model is used.

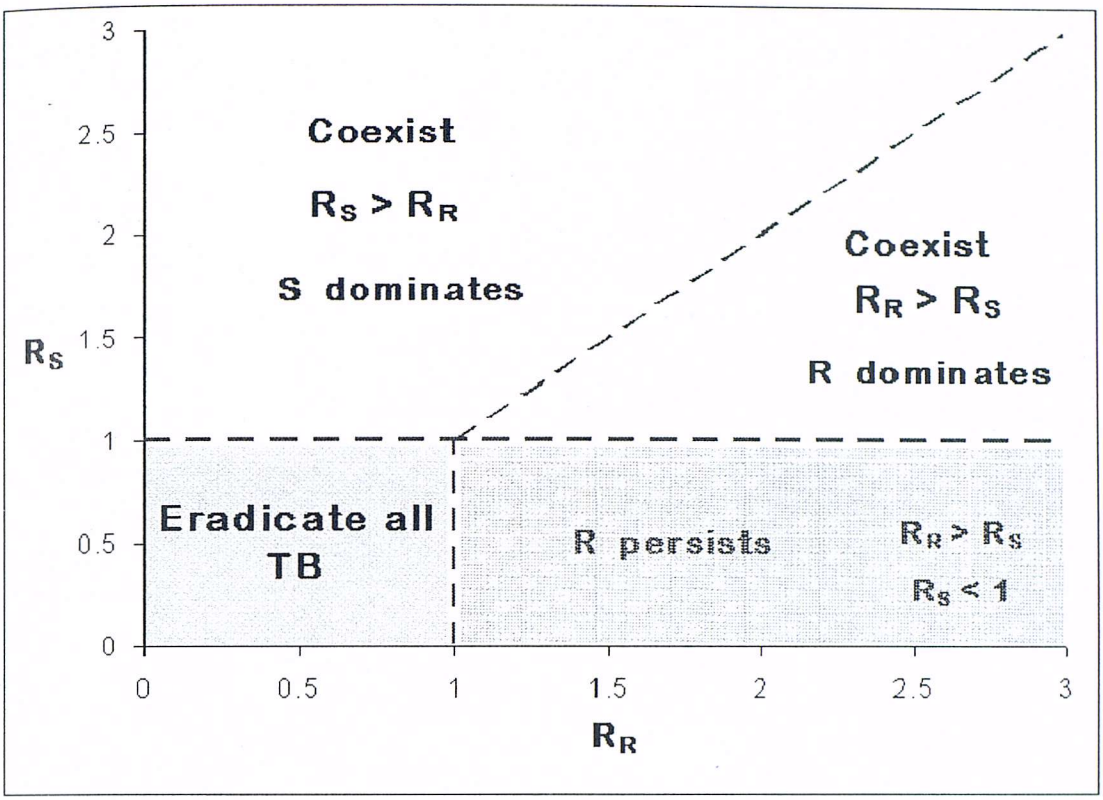


Figure 5-8. *The model outcomes depending upon R values.* The model outcomes for the two strain model, as described in Table **5-4** are illustrated. Three outcomes are possible under any given treatment strategy: eradication of all strains, persistence of only drug resistance, or coexistence of drug sensitive and resistant strains. When there is coexistence, drug sensitive strains will dominate the infectious pool when $R_S > R_R$, and resistant strains will dominate when $R_R > R_S$.

VI. A. Critical treatment rates for eradication all TB strains

Using the criteria defined above, the control parameters required to eradicate all strains in a TB epidemic can be determined. The threshold level of treatment (n) and treatment efficacy (Φ^C) required to eradicate all three TB strains is calculated.

Equation 5-17

A.

$$n\,\phi_S^c > \frac{c_S\left(i\,p + i\,\frac{(1-p)\,v}{\mu + \upsilon}\right) - (\mu + \mu_T + \kappa) + \frac{\kappa\,(w + \gamma\,i\,p\,\lambda_S)}{\mu + w + \gamma\,i\,p\,\lambda_S}}{(1 - \frac{(w + \gamma\,i\,p\,\lambda_S)}{\mu + w + \gamma\,i\,p\,\lambda_S})}$$

B.

$$n\,\phi_R^c > \frac{c_R\left(i\,p + i\,\frac{(1-p)\,v}{\mu + \upsilon}\right) - (\mu + \mu_T + \kappa) + \frac{\kappa\,(w + \gamma\,i\,p\,\lambda_R)}{\mu + w + \gamma\,i\,p\,\lambda_R}}{(1 - \frac{(w + \gamma\,i\,p\,\lambda_R)}{\mu + w + \gamma\,i\,p\,\lambda_R})}$$

C.

$$n\,\phi_M^c > \frac{c_M\left(i\,p + i\,\frac{(1-p)\,v}{\mu + \upsilon}\right) - (\mu + \mu_T + \kappa) + \frac{\kappa\,(w + \gamma\,i\,p\,\lambda_M)}{\mu + w + \gamma\,i\,p\,\lambda_M}}{(1 - \frac{(w + \gamma\,i\,p\,\lambda_M)}{\mu + w + \gamma\,i\,p\,\lambda_M})}$$

The relationship between notification rate and R value for each strain is illustrated in Figure 5-9. There exists a critical treatment rate at which the R value for each strain, assuming a given cure rate, equals 1. Increasing the treatment rate beyond this value brings R<1 and results in removal of this strain at equilibrium conditions. As the strains are assumed to be unlinked in this scenario, with no emergence of acquired resistance during treatment, this allows for the removal of a single strain from the population while other strains persist. In this scenario, the treatment cure rate for DS cases is set at 90%, for DR cases at 70%, and for MDR cases at 40%. At low treatment rates, the combination of treatment and cure rates is not sufficient to reach the threshold for R<1 for any of the strains, and all strains persist. As the treatment rate is increased, there is a range at which the combination of treatment and cure rates result in the removal of the sensitive strain, but allow for persistence of resistance. At yet higher

treatment rates, as shown in this example for above 96% of cases treated, both sensitive and resistant strains are removed. (Figure 5-9) At moderate treatment values, the outcome is likely to tend to coexistence of sensitive and resistant strains. In order to select for persistence of only resistance, there must be high enough treatment and cure rates in order to clear all sensitive strains, while low enough to allow for a continuing epidemic of resistance. Finally, in order to eradicate all strains, the highest levels of treatment and cure for all strains is required.

There exists a balance between the treatment and cure rates. Increasing either the treatment or cure rates reduces strain fitness. In terms of eradication criteria, a lower rate of treatment can be somewhat compensated for by a greater treatment efficacy. Shown in Figure 5-10 is the combination of treatment and cure rates such that $R_S = 1$. (Figure 5-10)

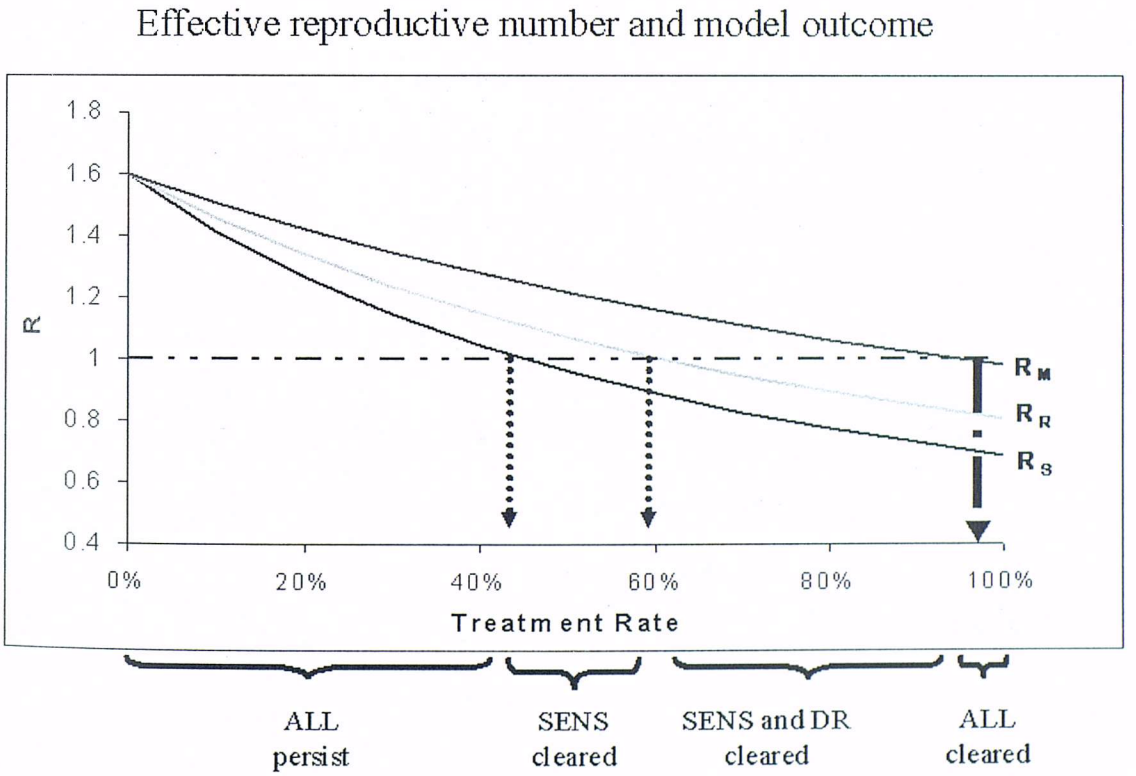


Figure 5-9. *Treatment rate and R value.* The proportion of cases receiving treatment (the treatment rate) is varied from 0% to 100% in simulations, and the resulting R value for each strain is shown. When there is no treatment (treatment rate = 0%), all strains have the same R value. Increasing the treatment rate reduces the R value of each strain, but less so for resistant strains, which have lower

treatment cure rates. The threshold R values required to eradicate S, R, and MDR strains are shown in terms of the treatment rate. In this simulation, for treatment rates from 0-40%, all strains persist; from 40-60% only sensitive is cleared; from 60-96% sensitive and singly resistant strains are cleared; and above 96% all strains are cleared.

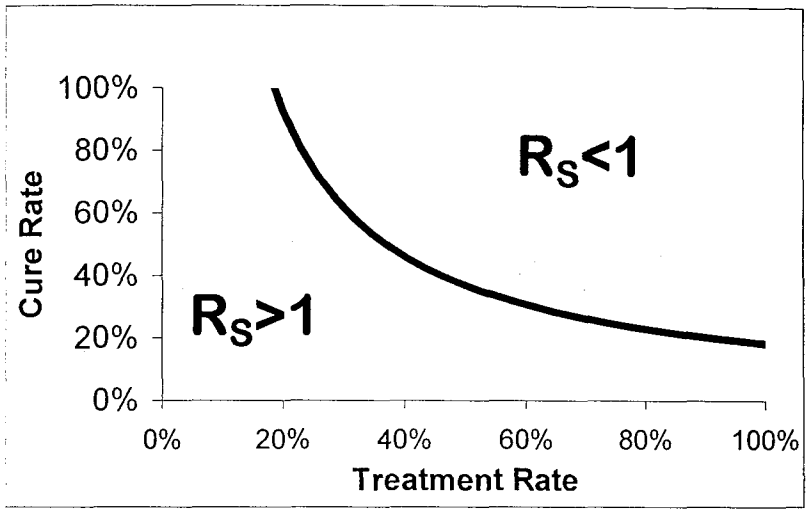


Figure 5-10. ***Balance between treatment and cure rate for eradication criteria.*** Increasing either the treatment or cure rate results in reduced R values for the strain. Shown is the combination of treatment and cu re rates such that $R_S = 1$. For treatment and cure rates above this threshold, $R_S < 1$ and the strain will be removed. Below this threshold, $R_S > 1$ and the strain will persist.

VI. B. Relative treatment efficacy for resistance

The relative treatment efficacy for drug resistance required to eradicate all TB strains can also be calculated. This measure can be useful in comparing the control strategies for treating drug sensitive and resistant cases. The formula below gives threshold values for the relative treatment efficacy of single (R) and multiple resistant (MDR) cases, compared to drug sensitive, required to eradicate the epidemic. (Equation 5-18) This is determined from the threshold $R_S = R_R = R_M = 1$, below which all strains in the population will be cleared.

Equation 5-18

$$\frac{\phi_M^c}{\phi_S^c} > \frac{\dfrac{c_M}{N}\left(i\,p + i\dfrac{(1-p)\,v}{\mu + \upsilon}\right) - (\mu + \mu_T + \kappa) + \dfrac{\kappa\,(w + \gamma\,i\,p\,\lambda_M)}{\mu + w + \gamma\,i\,p\,\lambda_M}}{\dfrac{c_S}{N}\left(i\,p + i\dfrac{(1-p)\,v}{\mu + \upsilon}\right) - (\mu + \mu_T + \kappa) + \dfrac{\kappa\,(w + \gamma\,i\,p\,\lambda_S)}{\mu + w + \gamma\,i\,p\,\lambda_S}} \quad \frac{\left(1 - \dfrac{(w + \gamma\,i\,p\,\lambda_S)}{\mu + w + \gamma\,i\,p\,\lambda_S}\right)}{\left(1 - \dfrac{(w + \gamma\,i\,p\,\lambda_M)}{\mu + w + \gamma\,i\,p\,\lambda_M}\right)}$$

These expressions for the threshold values of treatment required to eradicate a TB epidemic can be useful in designing and evaluating control programs. If control

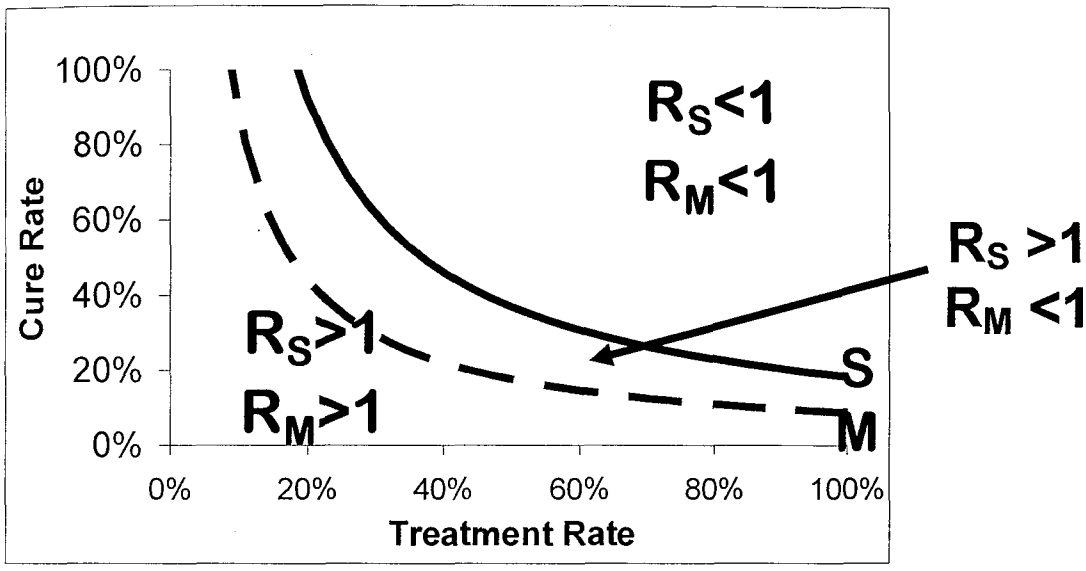
programs can achieve cure rates for resistant cases which are greater than those estimated here, than it is possible to prevent the scenarios depicted in the model in which resistance dominates at equilibrium. In this scenario, estimated critical relative treatment cure rate for MDR cases must be above 74% in order for MDR to have lower fitness than sensitive cases. This holds over all range notification rates. (Figure 5-11A) The critical values for the relative cure rates also depend upon the relative contact rate assumed, assumed in Figure 5-11A to be reduced by 10% for MDR cases. The relative cure rate for MDR cases required to keep $R_M<R_S$ is reduced with the MDR transmission rate, as shown in Figure 5-11B. Similarly, increasing the MDR fitness cost in transmission lowers the relative cure rate for MDR cases required such that MDR has lower overall fitness. (Figure 5-11)

These results can be compared with estimates for the relative cure rates of MDR cases on first line regimens. Resistant cases have been observed to have low cure rates on first line short course chemotherapy (SCC). Under effective DOTS SCC, cure rates for drug sensitive cases are near 90%. Cases with isoniazid resistance have cure rates reduced to 70-80%, while for MDR cases cure rates are at highest 60%. (Raviglione, Gupta et al. 2001) One study estimated the relative cure rates for resistant cases under DOTS SCC treatment in program conditions, and found that the cure rate was reduced in proportion to the number of drugs the case was resistant to. MDR cases had cure rates ranging from 6% to 59%. Even in regions with 100% DOTS implemented, and high cure rates for drug sensitive cases maintained, the cure rates for MDR cases was only 58% in Peru and 60% in Hong Kong. (Table 5-3) (Espinal, Kim et al. 2000)

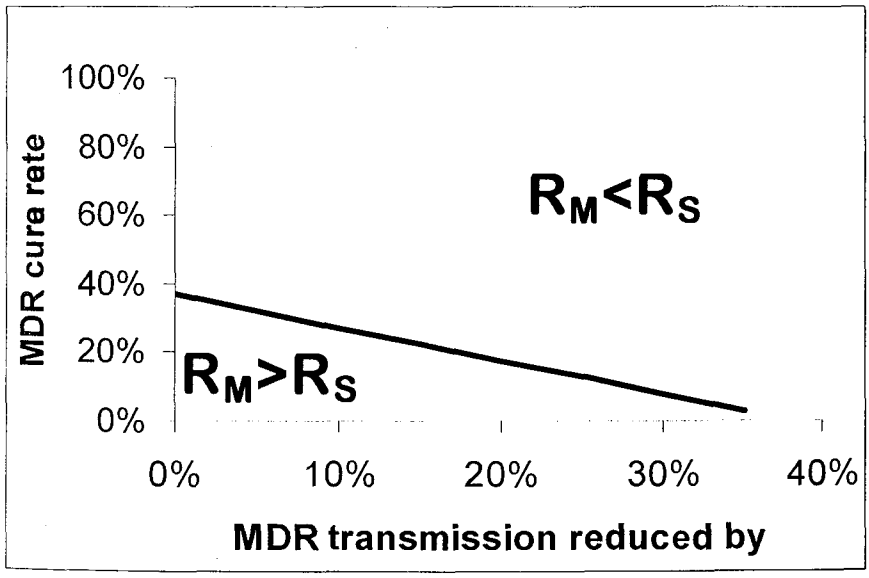
As illustrated in Figure 5-11C, if MDR has equal transmissibility as sensitive strains but reduced cure rates, then $R_M > R_S$. As the MDR transmission rate is reduced, it is possible for the MDR strain to have lower fitness than the sensitive strain, even with reduced cure rates. The greater the fitness cost in MDR transmission, the lower

the MDR cure rate required to keep the fitness of the MDR strain below that of the sensitive. (Figure 5-11C) Even with relative cure rates as high as 60%, MDR strains would need to have at least 15% reduced transmissibility to have $R_M<R_S$. This suggests that in most program conditions, MDR strains will have greater fitness unless they experience significantly reduced transmission.

A.



B.



C.

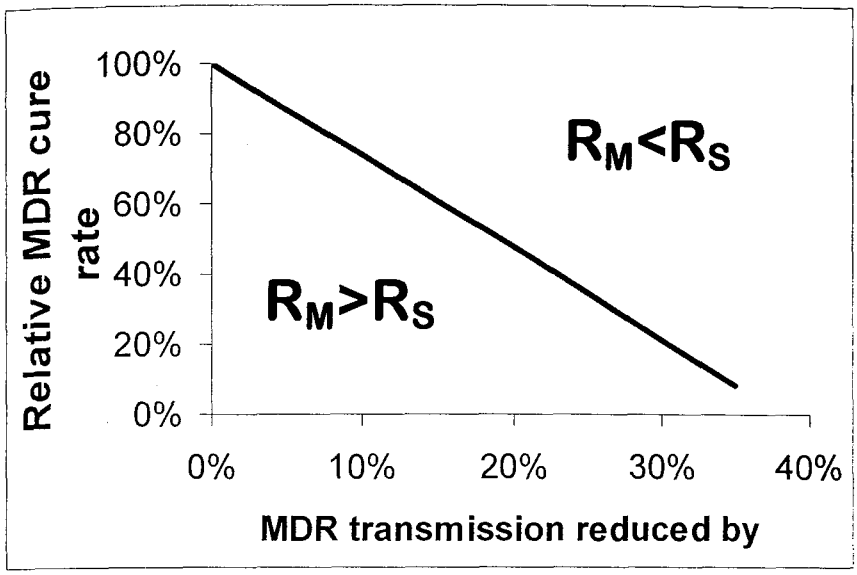


Figure 5-11. ***Critical treatment and cure rates for eradication of sensitive and MDR strains.***

A. Critical treatment and cure rates for eradication of drug sensitive (S) and MDR (M) strains are shown such that R_S or R_M, respectively, are equal to one. At treatment and/or cure rates above this line, $R_S > 1$ and $R_M > 1$, and both strains will persist. In between the two lines, $R_S < 1$ while $R_M > 1$, and the MDR strain will out compete the sensitive strain and dominate the population. For treatment and cure rates below both lines, both R_S and $R_M < 1$ and both strains will be removed at equilibrium. MDR cases are assumed to have a 20% reduced transmission rate compared to DS cases (f=20%)

B. The cure rate for MDR cases such that $R_M<R_S$. As the fitness cost of the MDR strain (f) is increased (by reducing the relative MDR transmission rate), a lower MDR cure rate is needed to maintain $R_M<R_S$. The cure rate for sensitive strains is set at 90%, and the treatment notification rate at 50%.

C. The relative cure rate for MDR cases compared to sensitive cases such that $R_M<R_S$. As the fitness cost of MDR cases (f, in transmission) is increased, a lower relative MDR cure rate is required in order to keep $R_M < R_S$. This relationship holds true over all treatment notification rates.

VII. Age Structure

As reviewed in Chapter 4, some previous models of TB have incorporated age structure. In terms of parameters, previous studies have suggested that the probability of progressing to primary disease, reactivation rate, proportion developing infectious disease, and the susceptibility to exogenous reinfection, self-cure rate, and the TB specific mortality rate can be considered to vary amongst age groups within a population. Studies modeling the natural history of TB epidemics have suggested the importance of including age structure and differentiating the relative disease risks by

age, in terms of understanding the progression of an epidemic within a population. These earlier modeling studies have not examined drug resistance, or considered the importance of including age structure on modeling and predictions of drug resistance. In this section, I briefly examine the importance of incorporating age structure into the model developed here.

VII. A. Age structured model

In order to consider the importance of age within our model, I incorporated age structure into the basic model, which includes one drug sensitive and one drug resistant strain. The assumptions regarding the basic model structure and parameter estimates are the same as developed within this chapter. The model is further defined with 5 year age classes, updated at the end of each year, with a constant birth rate into new susceptibles of age 0. Individual lifespan is set at 80 years, allowing only for TB-specific disease mortality to shorten an individual's lifespan.

As the clinical manifestations of tuberculosis disease depend on age, I assume some of the model parameters vary with age, based on survey of the literature (as reviewed in Chapters 2 and 3) as well as parameters used in other TB models (reviewed in Chapter 4). I review here the model parameters considered to vary by age within this section; the remaining parameters are assumed to be constant over the age classes.

The probability of progressing to primary disease (p) is considered to be higher in the young (age 15-24) and oldest age groups, perhaps as each may have reduced immunity or protection from infection. There tends to be a lower case rate between the ages of 0 – 14, as individuals infected during these ages have reduced progression to disease. In adolescents (age 15-24) there is a high rate of progression to disease, and clinical forms include either childhood or progressive primary tuberculosis. (Starke JR 1995; Donald PR 2000)

The proportion developing active pulmonary, infectious disease (i) is assumed to 50% in the age classes of 25 years and above. A lower proportion of infectious disease is assumed for the age classes of 0-25 years. Tuberculosis disease in the young is more likely than adult TB to be extrapulmonary (EPTB), or other forms, such as military TB. Those cases which are pulmonary (PTB) manifest fewer symptoms than adult cases, rarely producing sputum and usually culture and smear negative. (Wong, 1994; Donald PR 2000)

The reactivation rate (v) is considered to be higher in the oldest age class, which may overall experience lower immunity. The TB-associated mortality rate (u_T) as well overall mortality rates vary by age. Both the youngest and oldest age classes were assumed to have higher risk of disease-associated mortality, as suggested by references. The notification rate is assumed to vary slightly with age, with the youngest age classes assumed to have slightly lower notification rates. This is based on references suggesting difficulty of case notifications in children. (Wong, 1994; Donald PR 2000)

VII. B. Simulations

Simulations were started with equal numbers of all susceptibles in each age class, into which one infectious drug sensitive case in the age class of 35-39 years was added. The simulations were followed for 100 years. Following the change in age distribution of individuals and cases over time, showed that for the first 10-20 years, the population size remained near constant, and then began to decrease, as the disease prevalence increased. After approximately 40 years, the disease prevalence approached equilibrium. At this time, there was a decrease in the size of the older age classes, a moderate decrease in the middle age classes, and no decrease in the young age classes. This corresponds to the prevalence of disease in each class.

1. Low and high incidence scenarios

Next, I compared the age distribution of cases in scenarios of low and high disease incidence. In young age groups, the highest proportion of disease is caused by primary disease, under both scenarios. In comparison, the older age groups (approximately 50-80 years), contributed more of new incident cases in the low incidence (Figure 5-12A) than the high incidence scenario (Figure 5-12B). The highest proportion of disease was caused by reactivation disease in a low incidence scenario (Figure 5-12A), and primary disease in a high incidence scenario (Figure 5-12B). Each of these scenarios was run for the100 years and the final incidence rates are shown. The higher proportion of primary disease in scenario B reflects the higher levels of disease and primary transmission. In comparison, in scenario A, there is a higher proportion of reactivation disease in the older age classes, reflecting the relatively lower levels of disease and primary transmission.

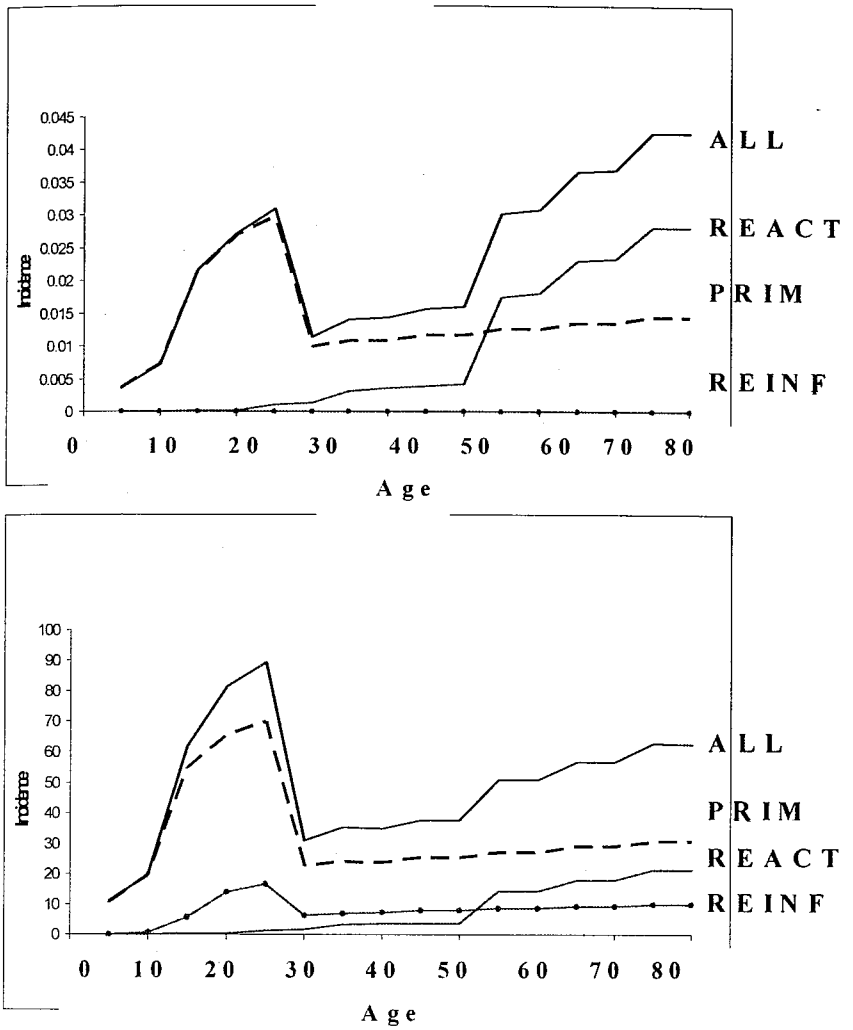


Figure 5-12. ***Incidence of disease across age groups, predicted by model.*** The incidence of disease by age was examined under a (A) low incidence and a (B) high incidence scenario. The incidence of primary disease (PRIM), reactivation disease (REACT), and disease due to exogenous reinfection (REINF) are shown under line which includes all disease (ALL). Incidence is given as new cases per 100,000.

VII. C. Drug resistance by age

When comparing the rates of resistance across age groups, shows that these are similar to drug sensitive cases. I next examined the incidence of drug resistance across the age groups. Again, I compared scenarios with either low or high incidence of disease. (Figure 5-13 A, B) In both scenarios, the trends in incidences of both DR and MDR cases appear similar to those of drug sensitive cases, across the age groups.

This brief consideration of age structure, suggests that, at least within the current model formulation and limited simulations, that inclusion of age structure does not significantly affect overall model considerations or predictions for resistance levels.

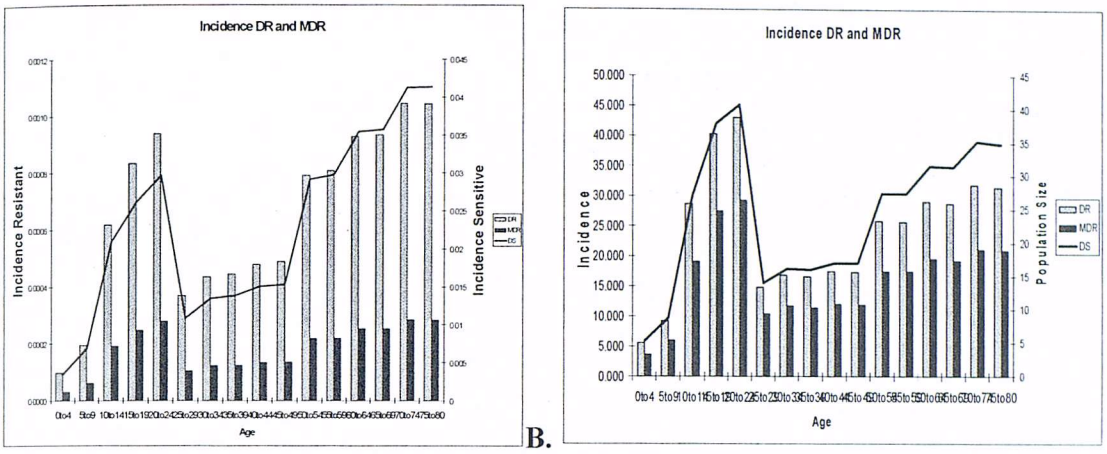


Figure 5-13. ***Incidence of drug resistance across age groups, predicted by model.*** The incidence of resistance by age was examined under a (A) low incidence and a (B) high incidence scenario. The incidence of drug sensitive, single (DR) and multiple drug resistant (MDR) cases, within model simulations. Incidence is given as new cases per 100,000.

1. Mycobnet surveillance on resistance

Next, I examined publicly available reports on the incidence of drug sensitive and resistant cases by broad age classes from a Mycobnet UK study, that includes reports of cases of active disease from centers across the UK from 1995-1999. Shown in Figure 5-14A are the notification rates of new active cases by year and age class. In Figure 5-14B are the numbers of these cases known to be INH resistant or MDR, again by age class. Comparing these two figures, it appears that the trends in resistance levels at the middle age classes are roughly comparable to the overall disease levels by age classes, with a peak in both the overall notification rates and the resistant cases in the 25-34 year age class. However, there appear to be fewer relative resistant cases found in the youngest (0-14) and oldest (65+) age classes. Comparing this to the above model simulations, we may expect that if there were no differences in the likelihood to develop resistance, that since there is a peak in the notification rates in the 65+ age class, there may also be a peak in resistance. These differences may be due number of

variables, including lower notification or detection of resistant case in these age classes, not accounting for other types of single (non-INH) drug resistance, and possibly limited or otherwise biased sampling.

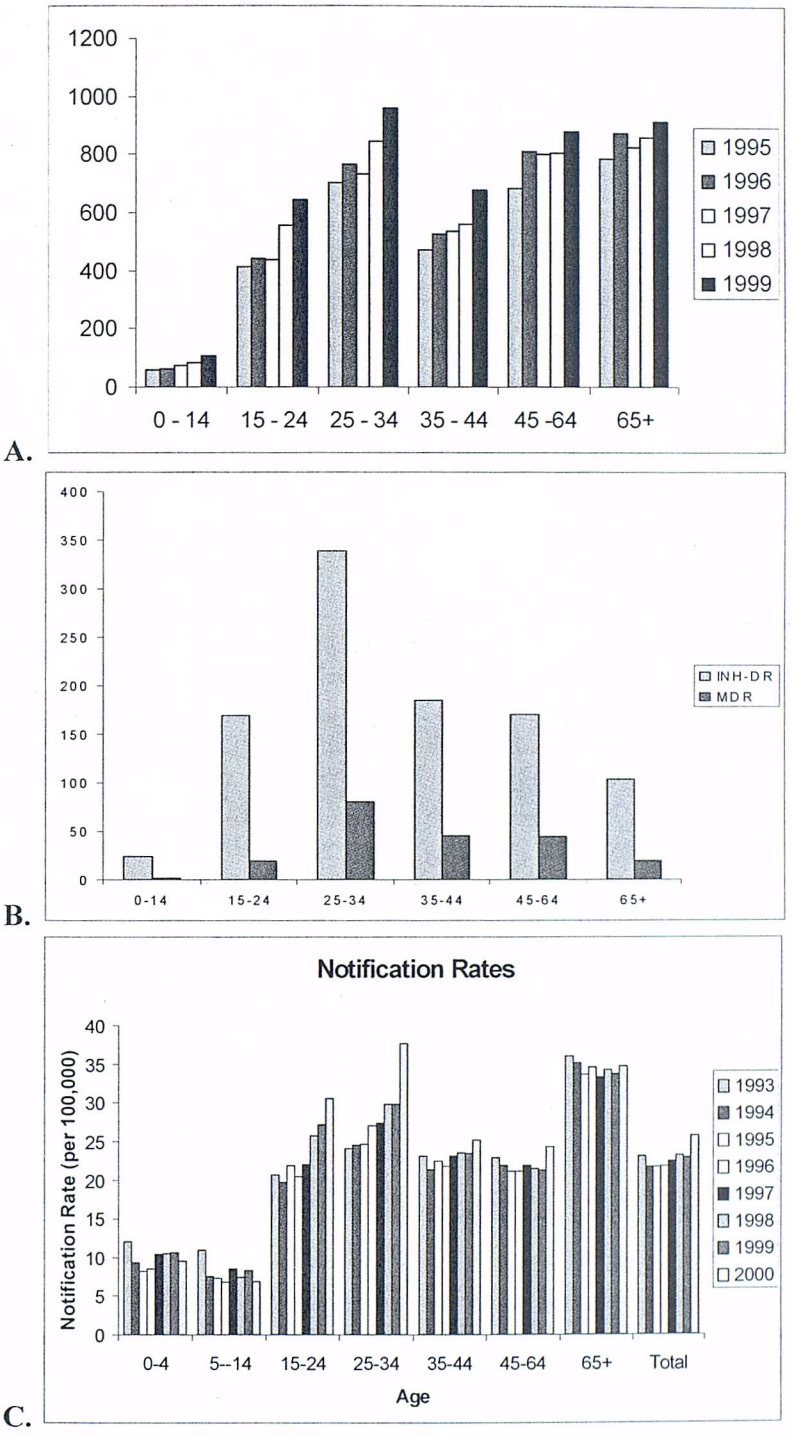


Figure 5-14. Notification rates of TB cases (A) and resistant cases (B) across age groups from PHLS Mycobnet survey in England and Wales from 1995-1999. (A) Initial M. tuberculosis isolates by age group, 1995-1999. (B) Isoniazid Resistance and Multi-Drug Resistance in Initial Isolates of M. tuberculosis, England

and Wales 1995 - 1999, by age group. (C) For comparison, from the WHO 2000 report, the Tuberculosis Notification Rates per 100,000 population, England and Wales, by age, 1993-2000.

VIII. Model parameter values

Parameter	Explanation	Estimated values	Source
N	*Population size*	*Held constant*	
λ	*Per capita rate of infection* $= c\ I/N$		
c	*Effective contact rate*	5-15 (average 10) infections/ infectious case - year	(Styblo, 1991), (Blower, McLean et al. 1995), (Porco and Blower 1998), (Dye and Williams 2000; Dye and Espinal 2001)
i	*Proportion infectious*	0.4 – 0.6	(Fishman and Elias 1998) (Davies 1998)
p	*Proportion infected progress to primary disease*	5-15%	(Sutherland, Svandova et al. 1982; Sutherland, Bleiker et al. 1983) (Vynnycky and Fine 1997)
v	*Rate latent infected progress to infectious disease*	0.03%	(Sutherland, Svandova et al. 1982; Sutherland, Bleiker et al. 1983), (Vynnycky and Fine 1997)
γ	*Reinfection parameter*	*Varied*	
κ	*Self-cure rate for DS cases*	10%	(Chaulet and Zidouni 1998) (Enarson and Hopewell 1998), Estimates in this chapter.
n	*Notification rate for sputum smear positive cases*		
ϕ_S	*treatment cure rate of DS cases*	*Developed: 70-95%, developing: 50-75%*	(WHO/IUATLD 2000; WHO 2003), (Chaulet and Zidouni 1998)
ϕ_R	*Treatment cure rate of single DR cases*	*Developed: 70-95%, developing: 50-75% (similar to DS cure rates)*	(Espinal, Kim et al. 2000)
ϕ_M	*Treatment cure rate of multiple DR cases*	*developed: 50-80% developing: 4-60% (reduced cure rates)*	(Espinal, Kim et al. 2000)

r	*Prob. of acquiring resistance mutation during drug treatment of DS case*	*Developed: low(5%) – high(10%) Developing: high (10-15%)*	(Chaulet and Zidouni 1998), (Dye and Espinal 2001)
μ	*Mortality rate (not TB)*	0.015	Average life expectancy 80 years
μ_T	*Mortality rate due to TB*	15-30 (mean 20)%	(Grzybowski 1991; Chaulet and Zidouni 1998; Davies 1998; Enarson and Hopewell 1998), Estimates in this chapter.
w	*relapse rate*	2-5%	(Chaulet and Zidouni 1998)

Table 5-5. ***Estimates of values for model parameters.*** Unless otherwise indicated, parameter values are given as rate per year. For control parameters (including the notification, cure, and acquired resistance rates) ranges of values found in developed and developing countries are given.

IX. Summary of Findings

IX. A. Model formulation

This model expands on previous in several respects. First, the interacting dynamics of sensitive and resistant cases within a tuberculosis epidemic are examined. Second, the emergence of acquired and transmission of primary drug resistance are incorporated, and the relative contribution of each to resistant sub epidemics examined. Third, the emergence of MDR is modeled in a two step process of the acquisition of resistance mutations.

Resistance is seen to emerge in a series of linked sub epidemics, from one founding drug sensitive case. This models the establishment of drug sensitive epidemics in the prechemotherapy era. The simulation begins with the introduction of drug treatment and follows the emergence of resistance, from the acquisition of mutation during treatment, and transmission as primary resistance.

IX. B. Mathematical definitions

Defining the basic (R_0) and effective (R) reproductive number in terms of model parameters provides insight into the model dynamics as well as the conditions under which resistance emerges and persists. The basic reproductive number (R_0) was derived by solving the system of equations at equilibrium for a model in which disease remained untreated. The effective reproductive number (R) was derived by finding the equilibrium solutions for a model which included the treatment of disease. The basic and effective reproductive numbers were derived for various versions of the model, including a model with one (drug sensitive), two (drug sensitive and resistant) and three (drug sensitive, singly resistant, MDR) strains, reinfection, and interacting strains. Both R_0 and R depend upon the effective contact rate, duration of infectiousness, and probability of progressing to active disease. Effective reproductive numbers were also derived for each strain, in which the contact rate and duration of infectiousness were strain-dependent, while the probability of progressing to disease was strain-independent. Treatment was seen to reduce the case duration of infectiousness, thus reducing the value of R.

Defining the effective reproductive number allowed for further examination of model dynamics. The relationship between model outcome at equilibrium and R values was examined. In terms of the balance between sensitive and resistance, three possible model outcomes exist: eradication of all strains, eradication of only the sensitive strain, or coexistence of sensitive and resistance. Resistance emerges when R_S and/or R_R are greater than one. If $R_S > 1$ but $R_R < 1$, then resistance will stay at a low level which is proportional to treated drug sensitive cases. The resistance prevalence will depend upon the proportion treated, as well as the cure and acquired resistance rates. When $R_R > 1$, an emerging resistance epidemic has the potential to continue in a self-sustaining cycle.

Critical treatment rates were defined for the eradication of one or more strains at equilibrium. Sensitive strains were removed at lower treatment rates than resistant strains. Higher treatment rates were required to reach threshold values such that both sensitive and resistant strains were cleared. There is also a balance between notification and cure rates in terms of eradication criteria: Increasing either reduces the strain fitness, increasing both simultaneously results in synergistic reductions in strain fitness, and reductions in one can also be compensated for by increasing the other.

These analyses apply to model equilibrium conditions, and thus not directly to predicting short-term changes in resistance prevalence as monitored in current global epidemics. However, several concepts are important in understanding TB resistance. These include the conditions for emergence of resistance, for competition between strains, and for sustaining an epidemic. First, competition between sensitive and resistant strains depends upon relative fitness. Second, resistance cannot be eradicated unless all sensitive strains are as well, as resistance will continue to emerge from the treatment of sensitive cases. Third, treatment has a differential impact on the fitness of each strain, with the implication that the conditions for eradication of each strain will differ, making it possible for one strain to succeed while another is removed under the same program. Lower treatment rates may allow for the clearance of sensitive strains, while allowing resistance to increase. Higher treatment rates are required to reduce prevalence of both sensitive and resistant strains.

6. Chapter 6

Dynamics of the Emergence and Transmission of Resistance

6. CHAPTER 6

DYNAMICS OF THE EMERGENCE AND TRANSMISSION OF RESISTANCE

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(2) Timescale
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(4) Reinfection

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I. Goals

I. A. To examine the dynamics of the emergence, transmission, and persistence of TB epidemics and resistance in model simulations.

I. B. To identify the conditions which most influence the emergence and spread of resistance within a population.

I. C. To explore the sensitivity of model predictions for disease and resistance levels to parameter estimates.

II. Abstract

The model for the emergence and transmission of resistant epidemics developed in the previous chapter is used here to consider the dynamics of resistance emergence, transmission, and persistence. In the absence of treatment, an epidemic emerges and reaches equilibrium levels of disease on a timescale of approximately 100 years. Introducing treatment reduces disease levels while selecting for resistance. Under poor control, with low cure rates, treating a greater proportion of cases increases the incidence of resistance. Under good control, with high cure rates for sensitive and resistant cases, increasing the treatment rates can contain and reduce the incidence of resistance. These results are most sensitive to the transmission rate of resistant strains. The greatest selection for the emergence and spread of resistance occurs under high treatment rates combined with low cure rates, when there is no fitness cost in resistance transmission. Under these conditions, increasing treatment coverage can increase the incidence of resistance as well as overall disease.

The sensitivity of the model predictions to parameter estimates is explored through Monte Carlo based simulations. In the absence of treatment, the model predictions for the disease incidence are most sensitive to parameter estimates for the contact rate, the proportion progressing to primary disease, and the reactivation rate.

When treatment is introduced, the disease incidence is most influenced by fixing the notification and cure rates. Similarly, the resistance prevalence is principally determined by the notification and cure rates. When the notification and cure rates are fixed, the resistance prevalence is most strongly influenced by the uncertainty in estimating the rate of acquired resistance, the fitness of the resistance strain, and the contact rate. Under good control, high treatment and cure rates limit the incidence of disease as well as prevalence of resistance. In comparison, under poor control, low treatment efficacy allows for higher levels of both disease and resistance to develop. These model predictions are most strongly determined by fixing the treatment parameters for the rate and efficacy of treatment.

III. Introduction

The burden of TB in a population is measured in several ways, including the incidence and prevalence of disease, mortality rates, and the force of infection. The commonly used statistics for reporting are the annual incidence or prevalence rate of cases per 100,000 population. Since 1993, global surveillance by the WHO has subdivided incidence rates into cases which are sputum smear positive or negative. (Raviglione, Dye et al. 1997; Dye, Watt et al. 2002) While the prevalence and incidence rates are routinely estimated in surveillance programs, there are only a few examples of estimates of the prevalence of infection and the annual risk of infection (ARI). The latter two require longitudinal estimates of the prevalence of infection within groups of a population over the course of a number of years. Such studies were done by Sutherland in the Netherlands; these landmark studies have provided the basis for current understanding of TB transmission rates. (Sutherland, Svandova et al. 1982) (Sutherland, Bleiker et al. 1983)

In this chapter, the burden of disease is measured as the incidence rate of new infectious cases per 100,000. For comparison, the prevalence of infection and prevalence rate of disease (as infectious cases per 100,000), are also shown in the first section. The resistance prevalence is given as the proportion of infectious cases with resistance. All case rates – for incidence, prevalence, and prevalence of resistance- refer to infectious cases.

Other mathematical models of tuberculosis epidemics have been formulated to consider the long term dynamics of tuberculosis epidemics. One has examined the natural history of tuberculosis over the course of the past century in England and Wales. (Vynnycky and Fine 1997) From this model, critical estimates of the dynamics of tuberculosis epidemics in low incidence regions have been made, including the importance of improvements in treatment, changing contact patterns, and the role of reinfection. (Vynnycky and Fine 1998; Vynnycky and Fine 1999; Vynnycky and Fine 2000; Vynnycky, Nagelkerke et al. 2001)

Other models have also examined the role of resistance in tuberculosis epidemics. One study simulated the transmission of resistant strains and used the model to estimate predicted levels of resistance. (Dye and Williams 2000) This model was also used to consider the impact of treatment with DOTS SCC on an epidemic of MDR, calculating critical rates for control of resistance. (Dye and Espinal 2001) Another model was used to consider the general dynamics of resistance and competition between sensitive and resistant strains in a population. (Blower, Porco et al. 1998; Blower and Gerberding 1998)

The study presented in this chapter adds several novel analyses. First, the emergence of a resistant epidemic from within a drug sensitive epidemic is examined. Second, the long term dynamics of resistance and the interaction with sensitive strains

are considered. Third, conditions for the emergence and spread of resistance are delineated. Fourth, detailed sensitivity analysis is performed to test the model predictions for emergence and spread of resistance to uncertainty in parameter estimates. Fifth, the key parameter uncertainties are identified within different control environments. Sixth, within the limits of this uncertainty in parameter estimates, the resistance prevalence in various control environments is predicted.

IV. Model Simulations

IV. A. Under no treatment

(1) Course of a typical epidemic

In the absence of treatment, introducing infectious TB cases into a susceptible population will allow for the emergence of an epidemic. The course of a typical TB epidemic, as used in model simulations throughout this chapter, is depicted in Figure 6-1. In this simulation, one active infectious case is introduced into a susceptible population of size $1x10^8$. Early in the epidemic, disease levels increase rapidly through ongoing cycles of transmission. (Figure 6-1A,B) The prevalence of infection continues to increase as the epidemic progresses. (Figure 6-1C) This reduces the number of susceptibles in the population, slowing the incidence of new cases until an equilibrium level is reached. (Figure 6-1)

(2) Timescale

The epidemic timescale, as shown here, is on the order of 100 years. This long timescale is a consequence of the slow rate of reactivation disease. The peak of endogenous disease may come several decades after the initial peak of primary disease, which reflects the rapid progression to disease early after infection. This

epidemic timescale is similar to that found in other models. (Blower, McLean et al. 1995; Blower, Small et al. 1996; Vynnycky and Fine 1997)

(3) Primary and reactivation disease

New incident cases are caused by either primary progression from recent infection or reactivation of an old infection. Early in the epidemic, a high proportion of disease is due to primary transmission. Over the course of an epidemic, the pool of latent infections accumulates, increasing the contribution of reactivation disease to new cases. As the epidemic progresses a greater proportion of cases are due to reactivation of latent infection. (Figure 6-1D)

(4) Reinfection

Introducing the possibility of reinfection of previously infected individuals into the model changes the course of an epidemic. As defined in the model, the reinfection parameter (γ) represents the susceptibility of previously infected individuals to reinfection and exogenous disease. Increasing the reinfection parameter resulted in an increase in incidence and prevalence of disease. As shown in Figure 6-2A, increasing the reinfection parameter results in a sharp increase in the incidence of exogenous disease and a slight increase in the incidence of primary disease. (Figure 6-2) The uncertainty over estimating the reinfection parameter has a large impact on the model outcome of disease incidence and prevalence. For the remainder of the simulations in this section, the reinfection parameter is set at 60%, in accordance with previous estimates. (See parameter estimates section of Chapter 4)

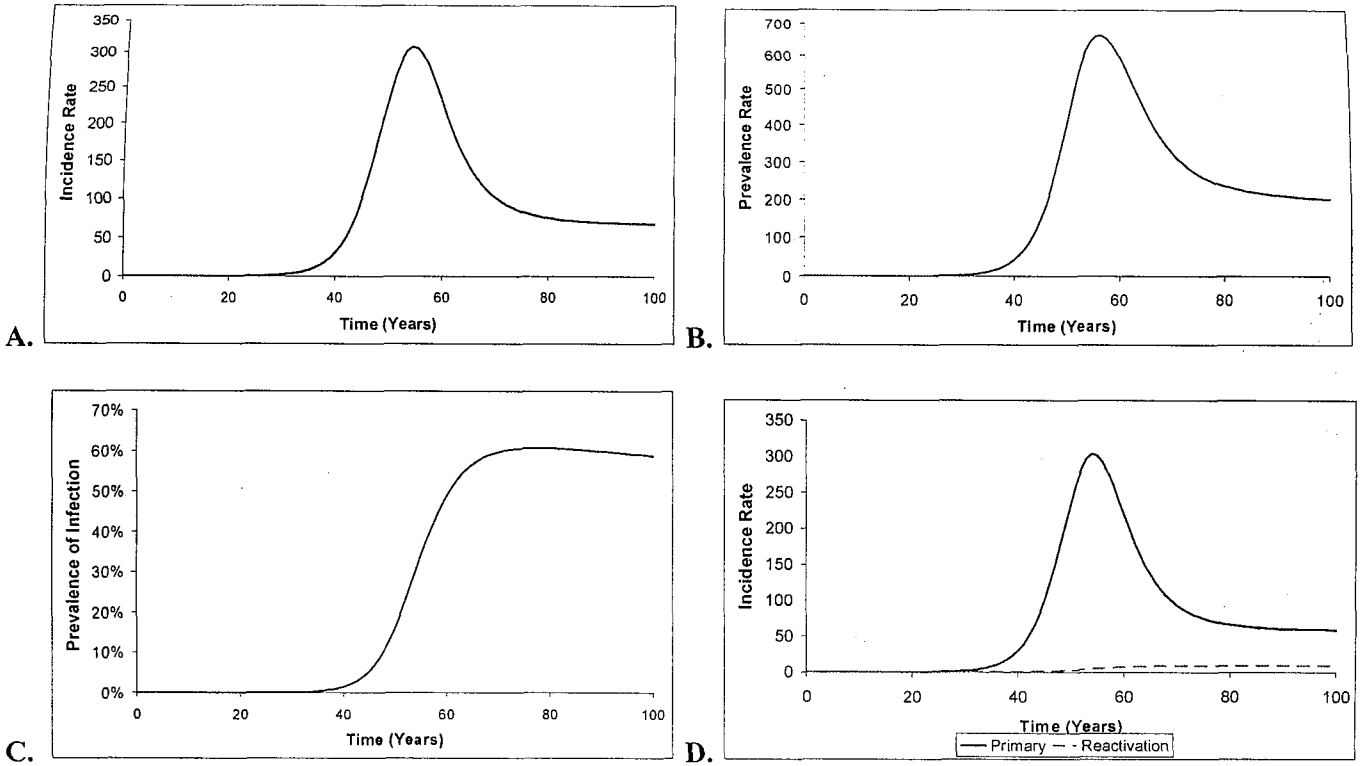


Figure 6-1. *The course of a typical TB epidemic.* The course of an epidemic is followed for 100 years. The simulation is begun by introducing one infectious case into a susceptible population. The incidence (A) and prevalence (B) of disease increase rapidly during the early phases of the epidemic, when there is a rapid supply of new susceptibles in the population. As a high proportion of the population is infected (C) few susceptibles remain and the incidence and prevalence of disease decline, reaching stable equilibrium levels. The incidence of primary disease accounts for most new cases early in the epidemic, while late in the epidemic the incidence of reactivation cases increases. (D) The incidence rate is shown as new infectious cases per 100,000 (A), and the prevalence rate as infectious cases per 100,000. (B) The prevalence of infection is shown as the proportion of the population with either latent infection, active disease, or recovered from disease. (C) The incidence rate of primary (solid line) and reactivation (dashed line) disease is shown as new infectious cases per 100,000. (D)

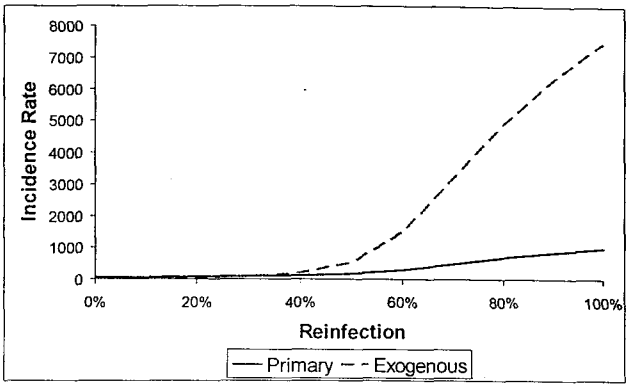


Figure 6-2. ***The impact of reinfection on the model predictions for disease levels.*** Increasing the susceptibility of previously infected individuals to reinfection increases the incidence of disease. Increasing the susceptibility to reinfection results in an increase in the incidence of first primary disease (solid line), but a greater increase in incidence of exogenous disease (dashed line). The reinfection parameter (γ), the model parameter reflecting the degree of susceptibility of previously infected individuals to reinfection disease, is varied in these simulations from 0% - 100%.

V. Treatment

(1) Reduces disease

Introducing a control program into the model simulations slows the emergence of an epidemic. Simulations were begun by introducing one drug sensitive infectious case into a susceptible population of size $1x10^8$, and followed for 200 years. In these simulations, it was assumed that the reinfection parameter was 80%. In years 0-100 of the simulations, disease is allowed to spread and reach endemic levels. In years 101-200 of the simulations, control is introduced. Introducing treatment reduces the incidence of disease. Introducing treatment, with a notification rate of 50%, reduces the incidence of disease. (Figure 6-8A) The treatment rate, equal to the proportion of incident cases receiving treatment, was varied from 0% to 100%. Increasing the treatment rate in these simulations reduces the incidence and prevalence of disease. (Figure 6-3B)

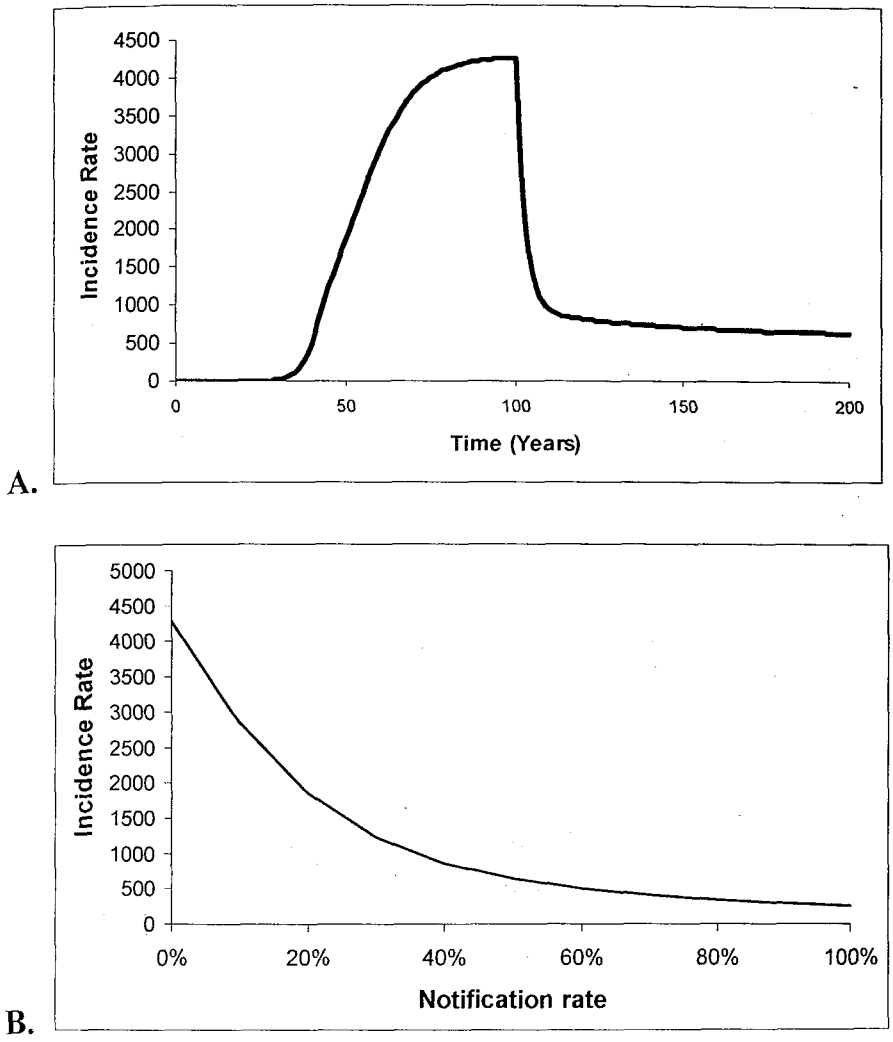


Figure 6-3. *The treatment of a higher proportion of cases reduces incidence of disease.*

A. Introducing treatment reduces the incidence of disease. Simulations were begun by introducing one infectious drug sensitive case into a susceptible population of size $1x10^8$, and run to equilibrium levels of disease in the absence of any treatment for years 0-100. From years 101-200, 50% of cases were treated. The cure rate for drug sensitive cases was set at 80%, for singly drug resistant cases at 60%, and for MDR cases at 40%. The reinfection parameter was assumed to be 80% in these simulations.

B. Higher notification rates reduce the incidence of disease. Simulations run as for A, with the proportion of cases receiving treatment (notification rate) during years 101-200 varied from 0% to 100%. Shown is the incidence rate of infectious cases per 100,000 at the end of the simulation.

(2) Emergence of resistance

Treatment also allows for the selection of resistance. Resistance emerges from the treatment of drug sensitive cases. As treatment is begun, resistance emerges in a series of linked sub-epidemics from within the initial drug sensitive epidemic. When

the conditions allow for resistance to both emerge and form a self-sustaining epidemic, resistance levels continue to increase. Simulations were begun by introducing one infectious drug sensitive case into a susceptible population of size $1x10^8$. Infectious cases received treatment at a rate of 50%. New cases of acquired resistance emerge from the treatment of drug sensitive cases, and continue a resistance epidemic through ongoing cycles of transmission of primary resistance. As treatment continues, the incidence of drug sensitive cases is reduced, while the incidence of DR and MDR cases increases. (Figure 6-4)

V. A. Transmission and selection for resistance

Several factors contribute the transmission and selection for resistance. The parameter most significantly associated with resistance is the treatment rate, with higher treatment rates correlated to higher resistance prevalence. (Figure 6-5) The extent of selection for resistance also depends on parameters for the cure rates, and resistance transmission rate. For a given treatment rate in these simulations, increasing the cure rate for resistant cases reduces the selection for resistance. (Figure 6-5A)

To explore the impact of varying the transmission rates of resistant strains, I first define the relative fitness of strains. The resistant strains are considered to have independent transmission rates, as reviewed in Chapter 2 and in the model developed in Chapter 5. Here, the fitness cost (f) of a MDR resistant strain is given as the transmission rate (c) relative to a wild type drug sensitive strain, as defined here:

$$f_{MDR} = 1 - \frac{c_{MDR}}{c_S}$$

Thus, if the transmission rate of the MDR strain was 80% that of the drug sensitive strain ($c_S = 0.8\ c_M$), then the MDR strain can be said to have 20% fitness cost ($f_{MDR} = 20\%$). This notation is used in the following simulations.

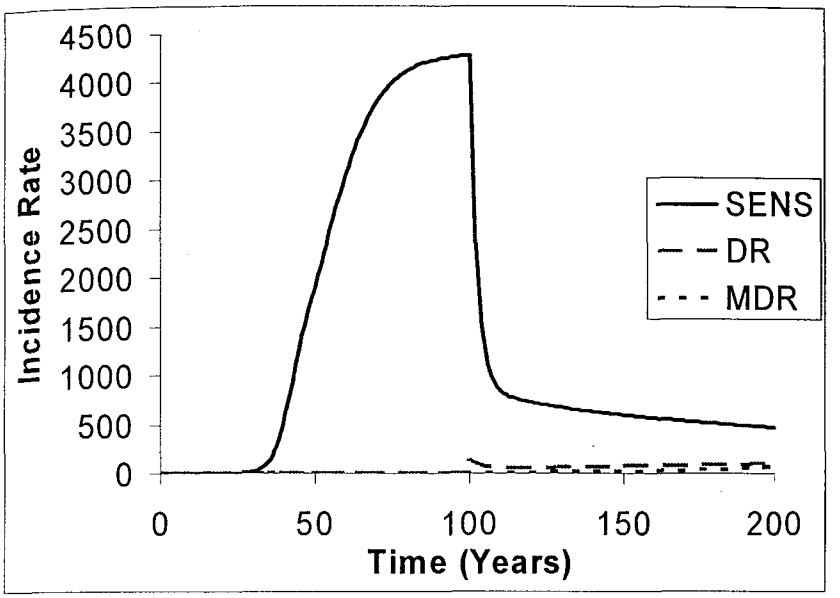
When resistant strains have fitness costs that reduce transmission (f_{MDR}) relative to drug sensitive strains, the selection for resistance is reduced. The greater the fitness cost of resistance, the slower the selection for resistance with increased notification rates. This is portrayed in two scenarios- under high or low cure rates. When there are high cure rates for both drug sensitive and resistant cases, the resistance remains at a low level (below 20%) even at high notification rates. Changes in the relative transmission rate of resistant strains have a small impact; reducing the relative MDR strain transmission by 50% (f_{MDR}=50%) reduces the resistance prevalence by 2%, when compared to estimates under equal transmission (f_{MDR}=0%). (Figure 6-5B)

When there are low cure rates for both drug sensitive and resistant cases, the relative fitness of the resistant strain has a greater influence on the selection for resistance. When the MDR strain has equal transmissibility as the drug sensitive (f_{MDR}= 0%), treating every case can result in a resistance prevalence as high as 80%. In comparison, when the MDR strain is 50% as transmissible (f_{MDR}=50%), treating all cases results in 35% resistance. (Figure 6-5C)

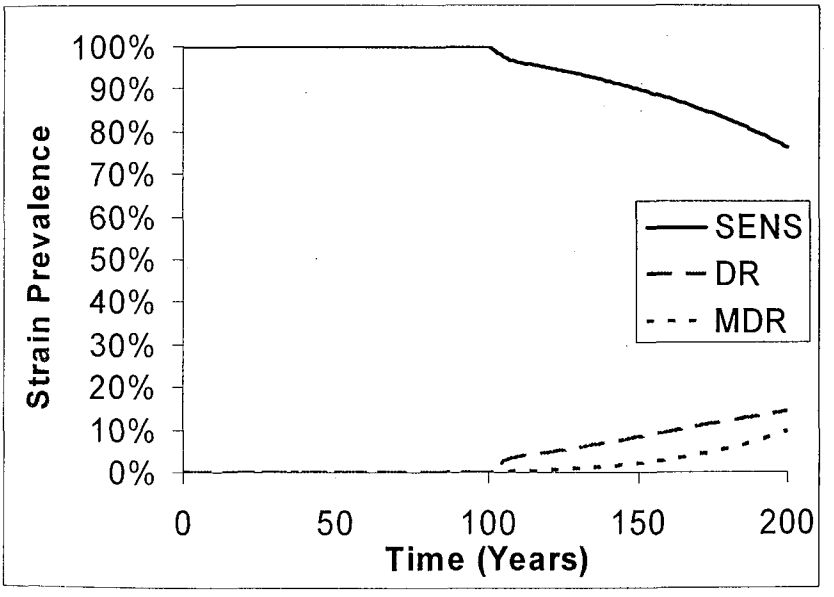
(3) Acquired and primary resistance

The relative proportion of resistant cases due which are either acquired during treatment or resulting from primary transmission reflects on the state of the resistant sub-epidemic. When resistance initially emerges within a drug sensitive epidemic, most of the resistant cases are those acquired during treatment. As the resistant cases transmit infection, the resistance epidemic grows. The relative proportion of resistant

cases due to primary transmission increases with the spread of the resistance epidemic. (Figure 6-6)



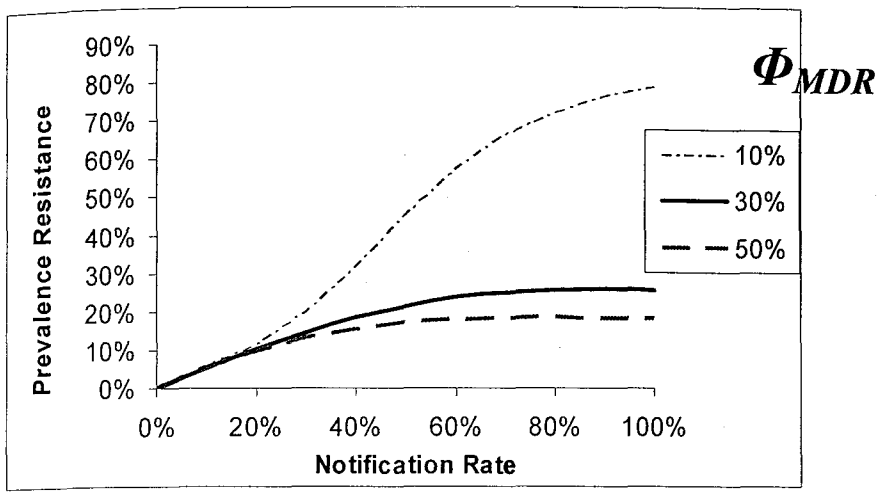
A.



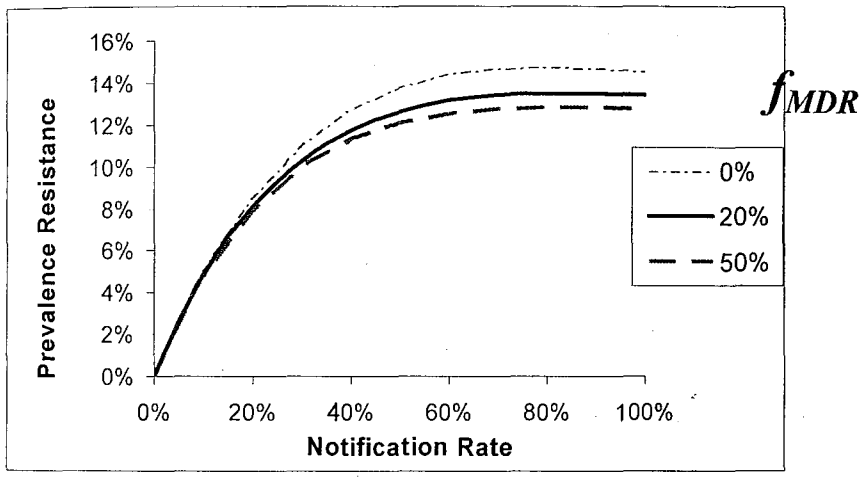
B.

Figure 6-4. ***Treatment selects for the emergence of resistance, which continues through ongoing cycles of transmission within the epidemic.*** The strain-specific incidence (A) and prevalence (B) are shown over the course of an epidemic under treatment. Treatment rapidly reduces the incidence of drug sensitive cases (DS), while allowing for the emergence of resistance. As treatment is continued, the incidence of resistant cases (singly resistant, DR, and MDR cases) increases. (A) Treatment also increases the prevalence of DR and MDR cases. (B) Simulations were begun by introducing one infectious drug sensitive case into a susceptible population of size $1x10^8$, and run to equilibrium levels of disease in the absence of any treatment for years 0-100. From years 101-200, treatment of 50% of cases was implemented. The cure rate for drug sensitive cases was set at 80%, for singly drug resistant cases at 60%, and for MDR cases at 40%.

A.



B.



C.

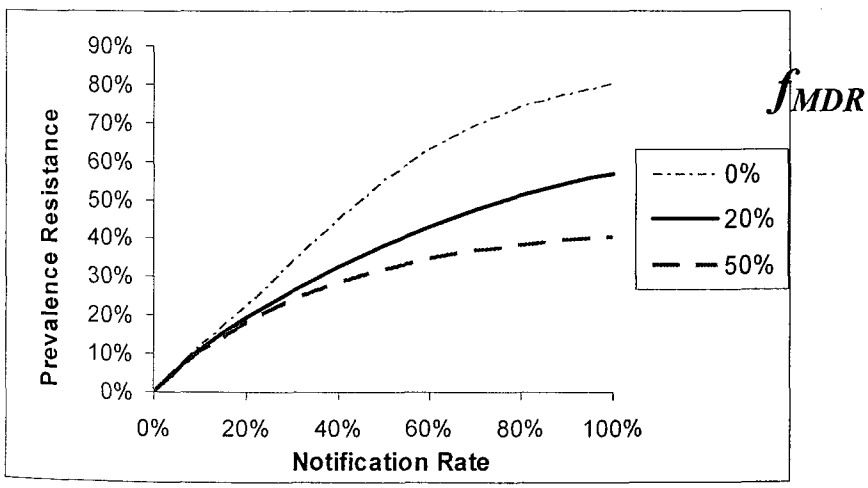


Figure 6-5. *Treatment selects for resistance. This selection depends most strongly on the resistant cure rate and fitness.*

A. Resistant cure rate varied. The cure rates for MDR cases was set at Φ_{MDR} = 10%, 30%, or 50%. At lower cure rates for MDR case, increasing the notification rate resulted in a greater increase in resistance prevalence.

B. Under good control. The fitness cost of resistance (f_{MDR}), representing the reduced transmission of MDR strain relative to DS, was set to 0%, 20%, or 50%. At greater fitness cost of MDR cases, increasing the notification rate resulted in less of an increase in resistance. The cure rate for drug sensitive cases was set at 80%, for singly drug resistant cases at 70%, and for MDR cases at 50%.

C. Under poor control. The cure rate for drug sensitive cases was set at 60%, for singly drug resistant cases at 40%, and for MDR cases at 20%. Increasing the fitness cost of resistance, reducing the transmission of the MDR strain, limited the selection for resistance with increased notification rates.

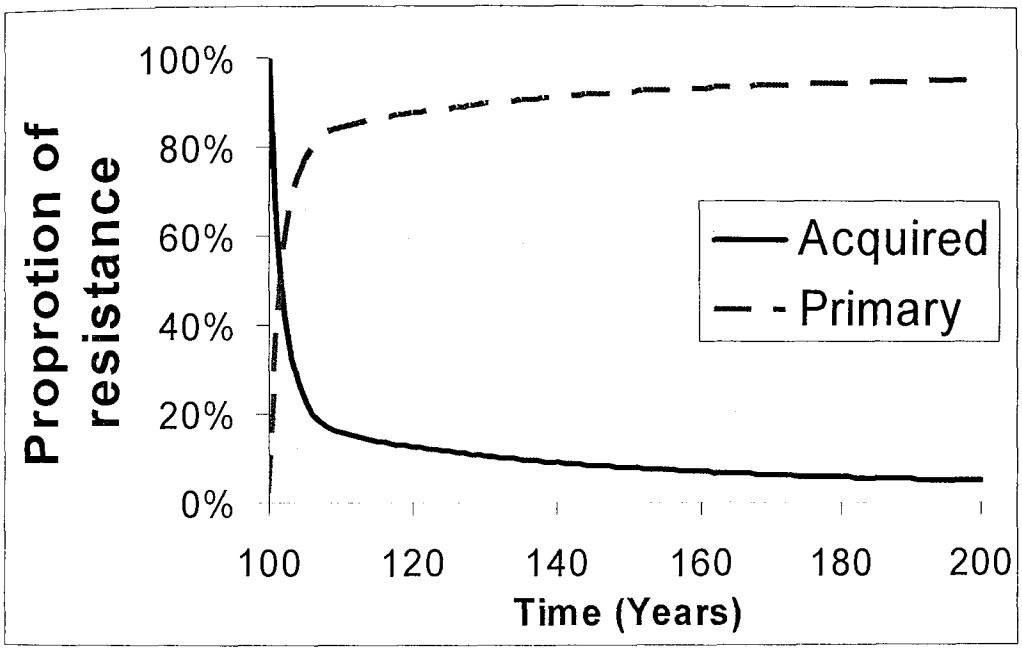
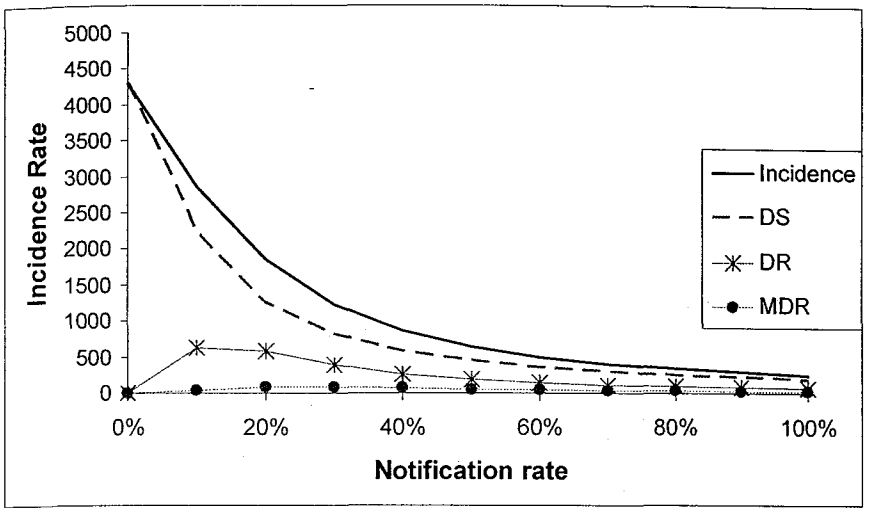
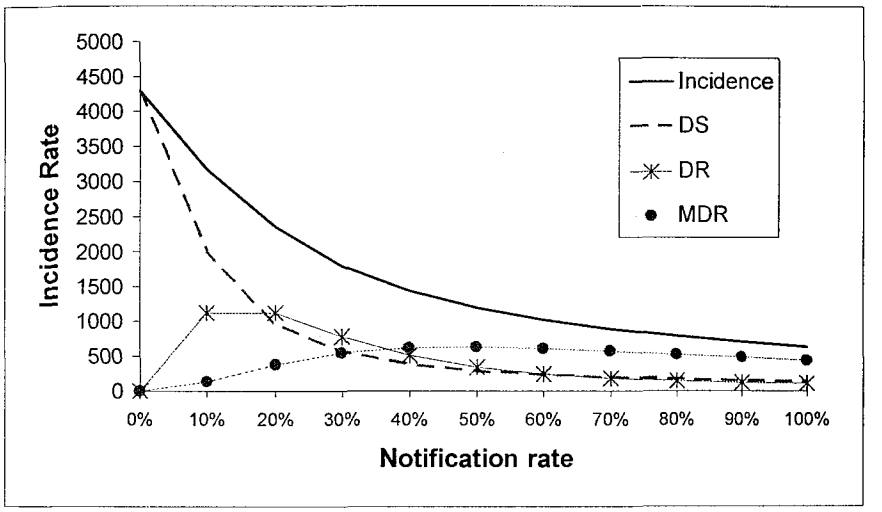


Figure 6-6. *The proportion of acquired and primary resistance over the course of an epidemic.* Early after the start of treatment at year 100, acquired resistance emerges from the treatment of drug sensitive cases. As the simulation continues these new cases of acquired resistance transmit primary resistance, and a greater proportion of resistant cases comes from primary transmission. Shown is the proportion of resistant cases for which resistance was either acquired during treatment (solid line) or from primary transmission (dashed line). Simulations were begun by introducing one drug sensitive case into a susceptible population of size $1x10^8$, and run to equilibrium in the absence of treatment for years 0-100. In year 101, treatment was introduced with a notification rate of 50%, a treatment cure rate for drug sensitive cases of 80%, for singly drug resistant cases of 60%, and for MDR cases of 40%. This treatment was maintained for years 101-200.



A.



B.

Figure 6-7. *Treatment selects for resistance, which can be reduced by high treatment and cure rates.* Increasing treatment rates can either control the emerging resistance epidemic or select for higher levels of resistance, depending upon the control conditions and relative fitness. Simulations were begun by introducing one infectious drug sensitive case into a susceptible population of size $1x10^8$, and run to equilibrium levels of disease in the absence of any treatment for years 0-100. The proportion of cases receiving treatment (notification rate) during years 101-200 was varied from 0% to 100%. The final incidence of infectious cases is shown as cases per 100,000.

A. **Under good control.** The treatment cure rate for drug sensitive cases is set at 80%, for singly drug resistant cases at 60%, and for MDR cases at 40%. Increasing the notification rate results in a decrease in incidence of disease (total incidence, solid line). This is mostly due to a reduction in incidence of drug sensitive cases (DS, dashed line). The incidence of singly resistant (DR) and MDR (MDR, circles) cases is increased by increasing the notification rate from 0% to 20%. Increasing the notification rate further results in a reduction in incidence of resistant cases.

B. **Under poor control.** The treatment cure rate for drug sensitive cases is set at 60%, for singly drug resistant cases at 40%, and for MDR cases at 20%. Increasing the notification rate results in a decrease in incidence of disease (total incidence, solid line). This is mostly due to a reduction in incidence of drug sensitive cases (DS, dashed line). The incidence of singly resistant (DR) cases is increased by increasing the notification rate from 0% to 20%. Increasing

the notification rate further reduces the incidence of DR cases. Increasing the notification rate results in an increase of MDR cases (MDR, circles).

(4) Conditions for emergence and spread resistance

As shown above, introducing treatment reduces the incidence of disease. In Figure 6-7 the affect of treatment on the incidence of each strain is shown. Increasing the treatment rate sharply reduces the incidence of drug sensitive cases. The impact of treatment on resistant strains is more complex and depends upon the efficacy of treatment for both sensitive and resistant cases. When treatment does not provide an effective cure for resistant cases, then higher rates of treatment will select for greater resistance. This is portrayed in an example of a simulation under poor control, in which both the sensitive and resistant cure rates are low. (Figure 6-7B)

When there is effective cure for both sensitive and resistant cases, increasing the treatment rate may result in reduced incidence of resistance. As shown in Figure 6-7, low rates of treatment allow for the emergence of resistance; as the treatment rate is increased from 0 to 30%, there is a sharp rise in the incidence of resistant cases. Further increases to the treatment rate result in a decline in resistance incidence. This reduction of resistance with higher treatment rates is a result of two combined processes. First, at high treatment and cure rates, more cases receive effective treatment and are rapidly cured, allowing less opportunity for acquired resistance to emerge. Second, the high notification rate allows for a greater proportion of new resistant cases to receive treatment, limiting the ongoing spread of resistance. (Figure 6-7A)

Under good control, high treatment and cure rates limit the emergence of resistance. Increasing the treatment rates can contain this emerging resistance. Under poor control, there is a greater opportunity for emergence as well as spread of resistance. With little effective cure for resistant cases, resistance increases with the

proportion treated. Increasing the treatment rates can exacerbate a resistance problem, increasing resistance prevalence as well as overall incidence.

VI. Sensitivity analysis

VI. A. Methods

In the previous section, the basic dynamics of the model were outlined. The direct relationship between several model variables—including the reinfection, resistance transmission, notification, and cure rates—and model outcome were explored. The analysis presented in this section expands on this sensitivity analysis by considering the relationship between each variable and model outcome in the context of varying all variables over plausible ranges for each individual variable. The correlation between variable and outcome is examined, over the space of possible combinations of parameter values, to find which parameters most influence each model outcome. The sensitivity scenarios presented in this section are compared in Table 6-1.

Sensitivity analysis was performed on a two-strain version of the model presented in chapter 5, with one sensitive and one resistant strain (as given in Appendix 1). The sensitivity of model predictions to parameter estimates is examined through Monte Carlo simulations. Simulations were run in C++ with RK4 method and a time step of one month. In each scenario given below, parameter values were varied simultaneously to give the range of possible parameter combinations. A normal distribution for parameters values was assumed, with mean and variance as given for each scenario. The resulting model outcomes were correlated to each parameter in turn. The influence of parameter variance on model outcome is portrayed with scatterplots showing the range of parameter values and the resulting

model outcomes. In order to further quantify the relative influence of each parameter on model outcome, the Pearson square coefficient (r^2) is calculated for the proportion of variance in the model outcome which can be explained by the variance in a given parameter. A higher coefficient (on a scale of 0 to 1) reflects a larger contribution to model outcome. (Full sensitivity analysis and scatterplots found in Appendix 3, summary of sensitivity analysis in Table 6-1)

For the purpose of this sensitivity analysis, the model parameters are classified into two general types: intrinsic and treatment parameters. The intrinsic parameters are those basic to the strain, transmission, and development of disease. These include the contact rate (c), proportion infectious (i), proportion progressing to primary disease (p), the mortality rate (μ_T), the self-cure rate (κ), the relapse rate (w), and the reinfection parameter (γ). The treatment parameters include the notification (nr) and cure rates (Φ), and the probability of acquiring resistance given treatment failure (r).

VI. B.Under no treatment

(1) Basic model, no reinfection

In scenarios 1-3, the sensitivity of model predictions in the absence of any treatment are examined. First, the model sensitivity was considered under a scenario where no treatment and no reinfection occur. The results of 1000 Monte Carlo simulations, run for 100 years in the absence of treatment, are shown in scenario 1A. The final incidence of disease ranged from 1-149 (average 81) cases per 100,000. (Simulation 1A) The final incidence of disease was most strongly influenced by the variation in the ECR (c), the proportion progressing to primary disease (p) and the endogenous reactivation rate (v). There was a positive correlation between each of

these input parameters and the model output of final incidence of disease. (Scenario 1)

(2) Reinfection sensitivity

In the second scenario, simulations were run as in scenario 1, but also included reinfection. The reinfection parameter (γ), representing the susceptibility of previously infected individuals to exogenous disease, was varied over a wide range from 0%-100%. The other intrinsic parameters were the same as for simulation 1. Introducing the possibility for reinfection increased the incidence of disease. In these simulations, the final incidence of disease ranged from 0-4403 (average 1140). The incidence of disease was most strongly influenced by variation in the reinfection parameter (γ). At low reinfection, the emergence of disease was limited to incidence rates below 1000 cases per 100,000, while higher reinfection allowed the opportunity for significantly higher levels of disease. The incidence of disease was also influenced by the parameters for the ECR and the proportion progressing to primary disease. (Scenario 2)

(3) Reinfection parameter fixed

In the third scenario, the reinfection parameter was held constant at 60%, the value used in subsequent simulations. The other intrinsic parameters were the same as for simulations 1 and 2. The final incidence of disease in these simulations ranged from 1-3217 (average 1540). The final incidence rate was, as in the previous two scenarios, most strongly influenced by the proportion progressing to primary disease and the ECR. (Scenario 3)

VI. C. Widely varied treatment

The model sensitivity is next examined in the presence of TB control, where a control program is defined as a set of notification, cure, and acquired resistance rates. In each scenario, simulations were begun by introducing one infectious drug sensitive case into a susceptible population of size $1x10^8$. For years 0-100, the epidemic was allowed to progress, in the absence of treatment, and reach endemic levels of disease. Treatment was introduced and maintained with a constant control program during years 101-150. In scenarios 4 and 5, the impact of treatment on model outcome is examined by widely varying the parameters delineating the control program.

In scenario 4, the reinfection parameter was also varied, in order to test the sensitivity of model behavior under treatment, and in particular on the emergence of resistance, to uncertainty in the reinfection parameter. The average final incidence rate was 355 (range 31 to 2899) cases per 100,000, with an average resistance prevalence of 17% (range 0%-96%). The final incidence of disease was most strongly affected by the uncertainty in the reinfection parameter (varied from γ=0%-100%), accounting for 50.8% of the variation in incidence. Higher reinfection parameters resulted in both greater and a wider range of incidence rates. The notification rate, also varied from 0%-100%, was the parameter with the second greatest influence on the incidence rate, accounting for 9.3% of the variance. Lower notification rates were correlated with a higher and greater range in incidence rates, while higher notification rates limited the incidence to low levels. (Scenario 4)

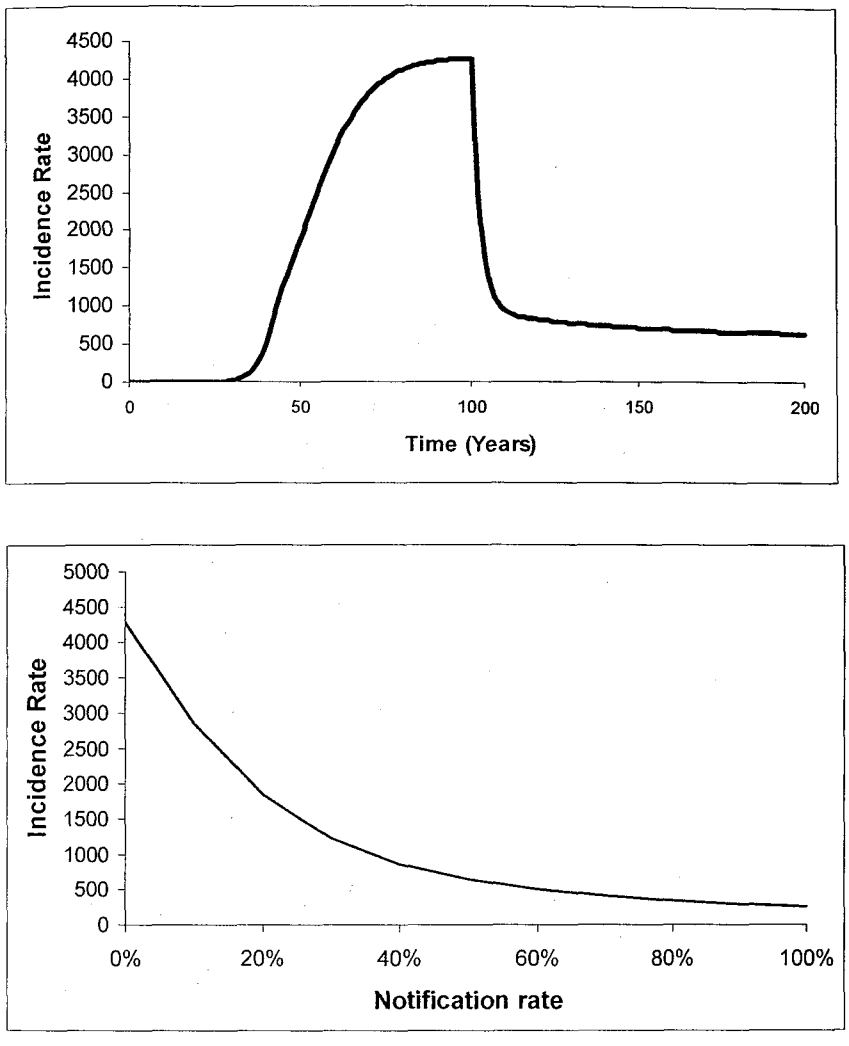


Figure 6-8. *The treatment of a higher proportion of cases reduces incidence of disease.*

A. Introducing treatment reduces the incidence of disease. Simulations were begun by introducing one infectious drug sensitive case into a susceptible population of size $1x10^8$, and run to equilibrium levels of disease in the absence of any treatment for years 0-100. From years 101-200, treatment of 50% of cases was implemented. The cure rate for drug sensitive cases was set at 80%, for singly drug resistant cases at 60%, and for MDR cases at 40%.

B. Simulations run as for A, with the proportion of cases receiving treatment (notification rate) during years 101-200 varied from 0% to 100%. Higher notification rates reduce the incidence of disease. Shown is the incidence rate of infectious cases per 100,000 at the end of the simulation.

Varying the reinfection parameter also had a significant impact on the final resistance prevalence. The uncertainty in the reinfection parameter had the largest impact on resistance, accounting for 23.5% of the variance. At higher reinfection parameters, there was a higher and greater range in resistance prevalence. The control parameters had the next greatest influence on the resistance prevalence; the DS cure

rate accounted for 12.7% of the variance, and the DR cure rates accounted for 13.3%, the acquired resistance rate for 9.1%. (Scenario 4)

In scenario 5, the reinfection parameter was set to 60%, as in the simulations following. The control parameters were widely varied, as in scenario 4. The average final incidence rate was 364 (range 63-1582) cases per 100,000, with an average resistance prevalence of 18% (range 0%-89%). With the reinfection parameter held constant, the greatest influence on the incidence rate came from varying the notification rate (from 0% to 100%), accounting for 37.6% of the variance. The final prevalence of resistance was most strongly influenced by the control parameters, followed by the transmission rate of the resistant strain. The DS cure rate accounted for 27.3% of the variance in final resistance prevalence, the DR cure rate for 20.4%, the acquired resistance rate for 14.2%, the notification rate for 9.8%, and the fitness of the resistance strain for 9.6%. (Scenario 5)

In scenario 6, the impact of widely varying the treatment rate was examined, while the cure rates were held fixed at high levels. The cure rate was set at 80% for DS cases and at 50% for DR cases. The average final incidence rate was 320 (range 23-1601) cases per 100,000, with an average resistance prevalence of 8% (range 0%-22%). The parameter with the greatest impact on the final incidence rate was the notification rate, which when varied from 0%-100% accounted for 52.4% of the variance in incidence. The parameters with the greatest influence on the final resistance prevalence were those for the control parameters and the fitness of the resistant strain. The uncertainty in the acquired resistance rate accounted for 57.8% of the variance in final resistance prevalence, the resistance transmission fitness cost for 20.4%, and the notification rate for 8.5%. (Scenario 6)

VI. D. Good control

In scenario 7, the model sensitivity is examined under treatment which is varied over a range representing 'good' control, with a high rate and efficacy of treatment. The notification rate was set at 75%, the cure rate for drug sensitive cases at 90%, and the cure rate for resistant cases at 60%. Simulations were run as in scenario 4, allowing the system to equilibrate in the absence of treatment for years 0-100, and receive a constant control program in years 101-150. The final incidence of disease in these simulations was an average of 157 (range 12-299) cases per 100,000, with an average final prevalence of resistance of 3% (range 1-5%). (Scenario 7A)

With the treatment parameters (aside from the rate of acquired resistance) already fixed, the model outcome for disease incidence was most affected by intrinsic parameters. The incidence of disease is most strongly affected by the ECR and the proportion progressing to primary disease. The final prevalence of resistance was most strongly influenced by the control parameter for the acquired resistance rate, as well as the intrinsic parameter for the resistance transmission rate. The uncertainty in the acquired resistance rate accounted for 54.9% of the variance in the resistance prevalence. Higher acquired resistance rates result in greater resistance prevalence. The fitness of the resistance strain, as measured by the relative transmission rate, accounted for 31.4% of the variance in the resistance prevalence. Increasing the fitness cost of resistance (by reducing the relative resistance transmission rate) limited the resistance prevalence to low levels. (Scenario 7) When the control program is fixed at high rates and efficacy of treatment, the greatest uncertainty in resistance prevalence comes from estimates for the acquired resistance rate, and the resistance transmission.

VI. E. Poor control

In scenario 8, the model sensitivity is examined under treatment which is varied over a range representing 'poor' control, with a low rate and efficacy of treatment. The notification rate was set at 50%, the cure rate for drug sensitive cases at 60%, and the cure rate for resistant cases at 30%. In addition, under poor control the rate of acquired resistance amongst treatment failures (r) was set to a higher value (and varied over a wider range) than under good control. Simulations were run as in scenario 4, allowing the system to equilibrate in the absence of treatment for years 0-100, and receive a constant control program in years 101-150. The final incidence of disease in these simulations was an average of 379 (range 44-791) cases per 100,000, while the final prevalence of resistance was an average of 37% (range 9%-67%). (Scenario 8A)

As in scenario 7, fixing the notification and cure rates leaves the model outcome for final disease incidence most strongly influenced by the intrinsic parameters of the ECR and the proportion progressing to primary disease. The final prevalence of resistance, again as in scenario 7, is most strongly influenced by the control parameter for the acquired resistance rate and the intrinsic parameter for the resistance transmission. These results appear to be common for fixing the notification and cure rates. However, the relative importance of the variation in the fitness of the resistant strain on the prevalence of resistance is much higher under poor control, while that of the acquired resistance rate is lower. (Scenario 8)

Table 6-1.

Simulation	Treatment parameters	Intrinsic parameters	Reinfection	Incidence rate (Cases per 100,000)			Prevalence of resistance (% of cases resistant)		
				Average	Min	Max	Average	Min	Max
1	None	Varied	None	81	0.7	149	-----	-----	-----
2	None	Varied	Varied	1140	0.1	4403	-----	-----	-----
3	None	Varied	Constant	1540	0.8	3217	-----	-----	-----
4	Control parameters varied	Varied	Varied	355	31	2899	17%	0%	96%
5	Control parameters varied	Varied	Constant	364	63	1582	18%	0%	89%
6	Notification rate varied over wide range	Varied	Constant	320	23	1601	8%	0%	22%
7	Good control	Varied	Constant	157	12	299	3%	1%	5%
8	Poor control	Varied	Constant	379	44	791	37%	9%	67%

Table 6-1. ***Comparison of Monte Carlo simulations.*** Each simulation consists of 1000 Monte Carlo runs of the model, with the intrinsic and control parameters varied between runs as given. The results of these simulations are shown as the final incidence of disease and prevalence of resistance.

VII. Summary of findings

VII. A. Basic model dynamics

The model predicts a course of a typical epidemic to occur on a timescale of 100 years. Early in the epidemic most cases are due to primary disease, while later in the epidemic there is an increase in endogenous reactivation disease. Increasing the reinfection parameter significantly increases disease levels.

VII. B. Conditions for emergence and spread of resistance

Introducing treatment reduces the incidence and prevalence of disease, but selects for resistance. The highest selection for resistance was seen under high notification rates combined with low cure rates. The continued spread of resistance to high levels depends upon the fitness of the resistance strain. Good control can limit the spread of resistance; increasing the treatment rates with high cure rates can reduce the incidence of resistance. Under poor control there is greater opportunity for the emergence as well as spread of resistance; increasing the treatment rates when there are low cure rates can exacerbate a resistance problem through increased selection as well as spread.

VII. C. Summary of sensitivity analysis

The sensitivity analysis showed that within the range of parameter uncertainty, the model can still make important conclusions. First, as expected, the model shows increasing treatment rates reduces disease and selects for resistance. Secondly, this depends upon the quality of treatment. Under good control, with high treatment rates and efficacy, there is a lower incidence of disease, as well as a limited emergence and spread of resistance. In comparison, under poor control there is higher incidence as

well as greater range in incidence, reflecting a greater opportunity for spread. Further, the low cure and higher acquired resistance rates under poor control result in higher resistance levels and greater range in resistance prevalence. Third, the greatest concern in terms of resistance emerging to high levels occurs in regions with poor control practices. The greatest potential for resistance, as well as the greatest model uncertainty in resistance prevalence, occurs under poor control. In order to address potentially serious emerging epidemics in such regions, the emergence of resistance should first be minimized through improving control practices, focusing first on improving quality of treatment and then expanding coverage.

In the absence of treatment, the model predictions for incidence of disease are most strongly influenced by parameters for the intrinsic dynamics of the epidemic. In particular, the reinfection parameter, ECR, proportion progressing to primary disease, and endogenous reactivation rate show the strongest impact on the final disease incidence. (Scenarios 1, 2, 3)

When the level of control is varied within a simulation, the control parameters have the greatest influence on model outcome for both disease and resistance. Under a widely varying level of TB control (Scenarios 5, 6), the control-specific parameters are most strongly correlated to model outcome. Once the control strategy has been defined and held constant throughout the simulation (Scenarios 7, 8), the parameters for intrinsic dynamics show increased influence on model outcome. When control strategy is fixed to high (good control, Scenario 7) or low (poor control, Scenario 8) levels, the prevalence of resistance is largely determined by variance in control parameters (and fitness cost), while disease prevalence is mostly affected by intrinsic parameters.

(1) Uncertainty in disease prevalence

In the absence of treatment, the incidence of disease is most strongly influenced by the ECR, the proportion progressing to primary disease, and the reactivation rate. (Scenario 1) When the reinfection parameter is varied, this parameter uncertainty has the greatest influence on final incidence of disease, with higher reinfection parameters resulting in increased incidence. (Scenario 2) When treatment is introduced, the incidence of disease is most influenced by the control parameters for the notification rate, the cure rate for drug sensitive cases, followed by the intrinsic parameters of the contact rate and proportion progressing to primary disease. (Scenario 4) This remains the same regardless of control environment. (Scenarios 5, 6)

VII. D. Uncertainty in resistance prevalence

The model uncertainty in resistance prevalence is most strongly determined by fixing the notification rate, followed by the cure rates for sensitive and resistant cases. (Scenarios 5, 6) When the control program is fixed at either high (good control, Scenario 7) or low (poor control, Scenario 8) levels, the prevalence of resistance is most influenced by the control parameter for the acquired resistance rate, and the intrinsic parameter for the resistance strain transmission. Under poor control, there is a greater opportunity for the emergence of high levels of resistance. Whether a high prevalence of resistance will emerge depends strongly upon the values for the acquired resistance rate and the fitness of the resistance strain. (Scenario 8) Thus, the model uncertainty in predicting resistance prevalence under a specified control program is almost entirely due to uncertainty in estimating the acquired resistance rate and resistance transmission, with a greater variance under poor levels of control.

VII. E. Uncertainty in key parameter estimates

(a) Resistance fitness

There is a great deal of uncertainty as to estimates for the fitness of resistance strains, as detailed in chapter 4 (section on parameter estimates). The sensitivity analysis presented in this section reveals the impact of this uncertainty in estimating the resistance transmission rate on model predictions for resistance prevalence. The variance in the resistant transmission rate was a primary cause of model uncertainty in resistance prevalence, in particular under poor control. (Scenario 8)

Under good control, the high treatment and cure rates limit the opportunity for the emergence of resistance. With lower emergence of resistance, there is less opportunity for spread to high levels, and varying the fitness of the resistant strain results in comparatively small fluctuations in resistance prevalence. (Scenario 7)

This understanding of the importance of the fitness of resistant strains can be used to inform control programs as to additional means to reduce resistance. The relative resistance transmission parameter, as defined in this model, reflects both contact as well as transmission. While the transmission rate is strain-dependent, the contact rate is believed to be strain-independent. However, interventions can target resistant cases in order to reduce their contact rates, thus reducing the overall resistance fitness. Where available, resources for active case detection can be focused on diagnosed MDR cases, finding and treating contacts of these cases. Increased isolation of these MDR cases can reduce contacts and thus transmission. This effectively occurs by sending MDR cases for treatment as specialized regional centers of excellence. (Mitnick, Bayona et al. 2003) (Farmer 2001) Finding and testing contacts of MDR cases can identify new latent infections or cases of disease for rapid treatment. (Loddenkemper, Sagebiel et al. 2002)

(b)Acquired resistance rate

As described in chapter 4 section on parameter estimates, there is significant uncertainty in estimating the probability of acquiring resistance during treatment. The sensitivity analysis of this section has shown that the model predictions for resistance prevalence are very sensitive to the uncertainty in the acquired resistance rate. This results in a wide range in resistance prevalence. Again, this is most significant under poor control, where there is the potential for the emergence of a high level of resistance. Under good control, does have significant impact on model predictions for resistance prevalence, but high case detection and cure limit the spread resistance to low levels.

The rate of acquired resistance amongst treatment failures is considered to be related to quality of treatment. A lower proportion of treatment failures acquire resistance under fully supervised and regulated DOTS programs. (Weis, Slocum et al. 1994) (Bastian, Rigouts et al. 2000) (Dye, Williams et al. 2002) Further, the probability of acquiring resistance has been correlated with errors in treatment, case management, and administration. (Mahmoudi and Iseman 1993) High rates of acquired resistance are seen in environments with poor control practices, less regulation, and other factors which might interfere with regular drug taking, such as poverty or war. (Iseman 1999) (Loddenkemper, Sagebiel et al. 2002) Improving these control practices can not only improve the treatment cure rates, but also reduce the probability of acquiring resistance. This underscores the importance, in regions with poor control, of first improving cure rates and reducing the probability of acquired resistance. Increasing treatment coverage before the quality of control is ensured can select for high levels of resistance. Once the probability of acquired resistance is reduced, there is less opportunity for the emergence of resistance, and treatment rates can be safely increased.

In order to restrict the model predictions for resistance prevalence, more information is needed regarding the quality of treatment. In localized settings, information regarding treatment practices can inform on the relative or probable rates of acquired resistance—and this can be used to narrow predictions for the emergence of resistance.

(c) Reinfection

As shown in the sensitivity analysis, varying the reinfection parameter significantly affects the incidence of disease in the model, both in absence (Scenario 2) and presence (Scenario 4) of treatment. Under treatment, the reinfection parameter also has a significant impact on the prevalence of resistance. Higher reinfection parameters allow the opportunity for emergence of greater levels of resistance. (Scenario 4) The reinfection parameter has the most influence on model outcomes in areas with high incidence and poor control.

VIII. Conclusions

1. **The model predicts realistic epidemic dynamics.**

2. **Resistance emerges under treatment, and the strength of selection for resistance is greatest under high notification combined with low cure rates, while continued spread of resistance depends heavily on the relative fitness of the resistant strain.**

3. **Sensitivity analyses showed that within range parameter uncertainty, the model continues to make useful predictions, and highlighted key parameters in representative scenarios.**

7. Chapter 7

Model for the control of resistant TB

7. CHAPTER 7

MODEL FOR THE CONTROL OF RESISTANT TB

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X. SUMMARY OF FINDINGS

I. Goals

I. A. To develop a model for control of TB epidemics that includes 1st and 2nd line treatment regimens.

I. B. To incorporate into this model realistic progression through the course of treatment, with case detection rate, time to detection and duration of treatment.

I. C. To include varying treatment efficacy by resistance status, and acquired resistance emerging during the course of treatment, depending on regimen and strain.

II. Abstract

In this chapter, the model presented for the emergence and transmission of resistance is expanded. The components of the model related to treatment are further developed, while the rest of the model remains the same as that outlined in chapter 4. Several implications of the formulation and parameterization of this model are explored. A sensitivity analysis of model results regarding the impact of control strategies, presented in the subsequent two chapters, is described. The duration of infectiousness (DOI) is defined by strain and treatment regimen. These definitions are used to examine the impact of treatment, at different rates and efficacy, on case DOI.

In addition, the importance of treatment times is examined. Treatment waiting times for detection and treatment are included. Each treatment course is divided into separate phases during which there are characteristic levels of case infectiousness, intensity of treatment, and probability of acquiring resistance.

III.Methods

III. A. Simulations

A compartmental deterministic model was developed, defined by the set of ordinary differential equations shown in Appendix 2. These set of equations were

solved in Berkeley Madonna using Runge Kutta 4 method with a timestep of 0.01 month. The model parameters are defined in Table 7-1 and Appendix 2.

III. B. Formulating the model

Individuals are born into the population susceptible (X). Susceptible individuals are infected at a rate equal to the force of infection (λ) and either progress directly to infectious disease (sensitive, I_S, resistant, I_R, or MDR, I_M,) or enter a latent state (L_S, L_R or L_M) from which there is a slow rate of reactivation. The force of infection is defined as the effective contact rate multiplied by the proportion of infectious individuals in the population. The effective contact rate (c_S , c_R, c_M) is defined here as the probability of contact and transmission of infection between an infectious and a susceptible individual. The force of infection is defined separately for sensitive (λ_S) and resistant (λ_R or λ_M) strains, and is given by:

$$\lambda_S = c_S \frac{(I_S + T_S^i + P_S)}{N} \quad , \quad \lambda_R = c_R \frac{(I_R + T_R^i + P_R)}{N}, \lambda_M = c_M \frac{(I_M + T_M^i + P_M)}{N}$$

A proportion of new infectious cases, equal to the case detection rate (CDR), are detected. The CDR is deceptively referred to as a rate (and not a proportion) in adapting the terminology of national surveillance definitions. After a time to case detection (t_{CD}) a proportion (CDR) of new infectious cases are detected and begin a course of treatment. The remaining (1-CDR) of new infectious cases are considered undetected (U_S, U_R or U_M) and receive no treatment.

Cases receiving treatment (T_S, T_R or T_M) are further subdivided according to treatment phase and regimen, as well as whether they are infectious. During the initial stages of treatment, effective treatment will rapidly reduce the patient's mycobacterial load, and will leave the patient non-infectious. Infectious patients (T_S^i, T_R^i, T_M^i) can transmit infection, while those rendered non-infectious during a course of treatment

(T_S^{ni}, T_R^{ni}, T_M^{ni}) continue the course of treatment and may retain symptoms (including an increased mortality rate) but cannot transmit disease. After a course of treatment, patients who have become non-infectious are cured of disease (C_S, C_R or C_M), while those remaining chronically infectious move into a class for previously treated patients (P_S, P_R or P_M).

IV. Course of treatment

IV. A. Mycobacterial numbers over course of treatment (theory):

The course of treatment is modeled here as the progression through distinct phases of a treatment regimen. The treatment course consists of an initial two week phase, followed by phases two and three, each lasting three months and together representing the main part of short course chemotherapy (SCC). The fourth phase of treatment also lasts three months and represents an extension phase.

A theoretical representation of the changes in within-host mycobacterial numbers over the course of treatment, according to the timing of treatment phases defined in the model, is presented in the figure below. Two scenarios are portrayed: one in which the patient remains chronically infectious and treatment fails (chronic), and one in which the bacterial infection is cleared and the patient is cured (cured). (Figure 7-1)

When the patient is cured by effective treatment, there is a rapid decline in mycobacterial load in phase 1. During this initial phase of treatment, effective therapy is thought to rapidly decrease the patient's mycobacterial load, corresponding to a reduction in symptoms and possibly a shift to be non-infectious, as the patient emits fewer viable mycobacteria in sputum. This is depicted in figure 6.1 as a rapid reduction in bacterial load below a threshold where the patient is no longer infectious. Treatment

is continued after this initial phase, even if symptoms have abated, to target the remaining mycobacterial population. In phases 2 through 4, treatment continues to clear the remaining bacteria in the patient. The decline in the bacterial load is modeled with two exponentials: a fast exponential decline in the first phase, followed by a slower exponential decline in the remaining phases of treatment. (Figure 7-1)

As direct evidence for typical mycobacterial numbers in human patients, or the change in these numbers over the course of treatment, are not available, these estimates reflect current qualitative understanding of these processes. The rapid reduction in symptoms and conversion of sputum test in the first month of treatment is thought to correlate with a clearance of a large proportion of the host's bacterial population. It is suggested that only a small bacterial population remains after this initial phase, and that possibly these bacteria are more difficult to clear either because of location, or some phenotypic resistance to drug action such as a slowed growth rate or dormancy. The treatment course must be extended past the initial phase in order to remove these remaining bacteria. (Dickinson and Mitchison 1976; Allen B.W 1992; Bloom 1994; Gangardharam and Jenkins 1997; Mitchison 2000)

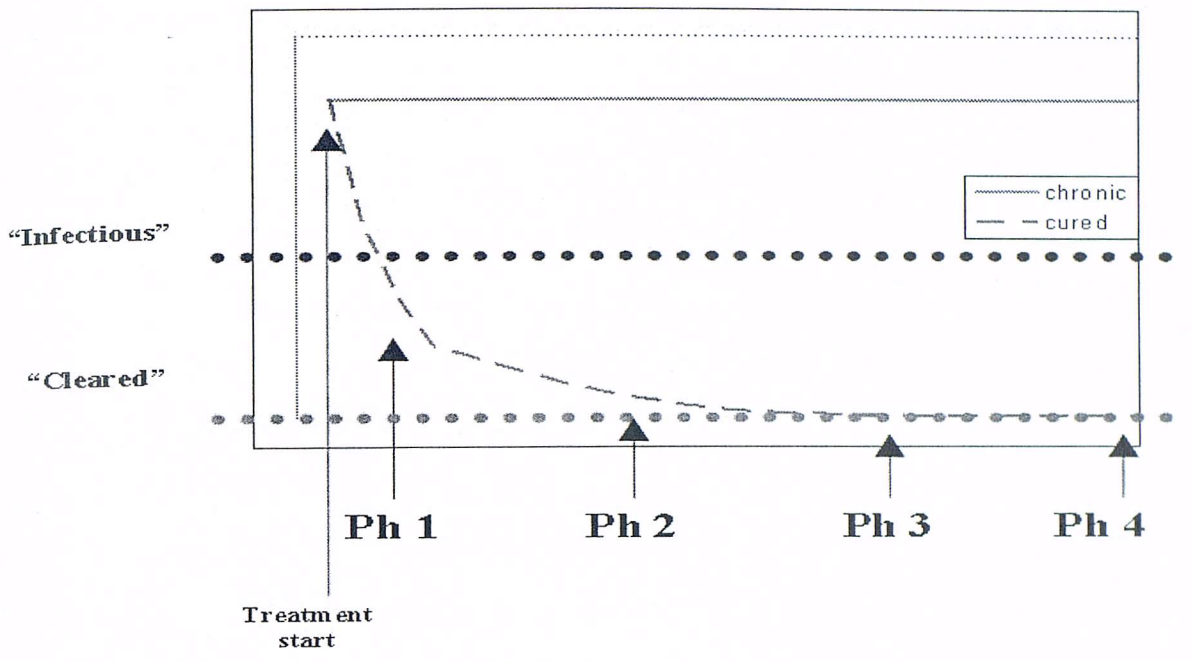


Figure 7-1. ***Mycobacterial load versus treatment time.***

Representation of the speculated changes in mycobacterial numbers within a patient over the course of treatment. The relative mycobacterial numbers is followed from the start of treatment through phases 1 to 4 (Ph1 – Ph4) of a treatment course. A patient failing treatment remains chronically infectious and maintains a high mycobacterial load (—). A patient effectively cured by treatment (- - -) is thought to experience a rapid decline in mycobacterial numbers in phase 1, followed by a more gradual decline in phases 2 – 4. Theoretical thresholds for the mycobacterial load required for case infectiousness and disease clearance are shown.

IV. B. Treatment patterns

Three treatment regimens are included in the model: a first line (regimen A) effective for drug sensitive (DS) cases, a second line (regimen B) for singly drug resistant (DR) cases, and a second line (regimen C) for multiple drug resistant (MDR) cases. First-line treatment reflects the typical SCC regimen of four drugs given for six months, followed by an extension phase of two drugs taken for three months. Second-line treatment reflects a specialized regimen given to a patient found to have drug resistance, and lasts longer. In the model, both DR and MDR cases receive an extended final phase of treatment.

Possible treatment scenarios include the treatment of a drug sensitive case with regimen A and the treatment of a drug resistant case with regimen A, B or C. The cure rates are highest for treatment of sensitive cases with regimen A, followed by singly resistant cases with regimen B, and MDR cases with regimen C. The treatment of a singly resistant case with regimen A is considered to have low effectiveness, as is the treatment of a MDR case with either regimen A or B.

IV. C. Model treatment course

A proportion of treated cases, equal to the cure rate (Φ), are effectively cured during a course of treatment. These cases become non-infectious after the first phase of treatment (T^{ni}). The remaining cases, ($1-\Phi$), continue through the course of treatment remaining infectious (T^{i}) and then move into a class for previously treated (infectious) individuals (P).

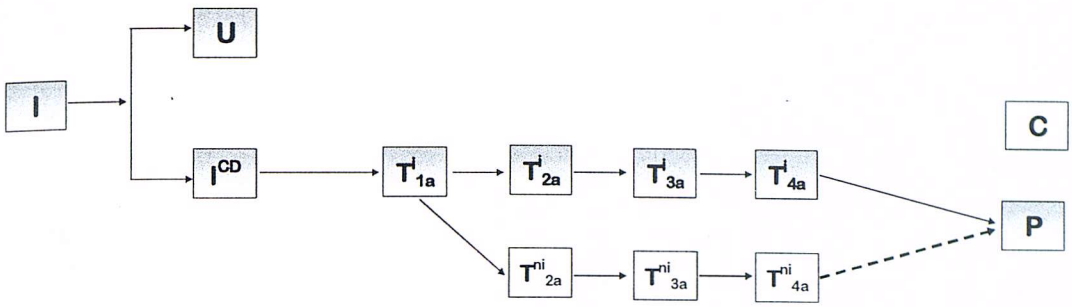


Figure 7-2. ***The model for treatment of DS cases.***
New incident cases (I) are either detected (I^{CD}) and begin a course of treatment (T) or remain undetected (U). Cases may be cured (C) through self-cure or treatment, or may fail a course of treatment and remain infectious (P). Darkened boxes represent infectious classes, while clear boxes represent non-infectious classes. The treated classes (T) are subdivided according to both treatment time (phases 1-4 shown in subscript) and infectious status (infectious (i) or non-infectious (ni) shown in superscript). A proportion of treated cases are cured and rendered non-infectious after the first phase of treatment: after completing the treatment regimen most of these cases (in T^{ni}) are transferred to the cured class (C), while a small proportion will relapse to disease (and enter P).

IV. D. Model for acquired resistance

Acquired resistance is modeled by allowing a proportion of treated DS cases which fail treatment to become resistant cases. A proportion (r_1) of DS cases not cured by treatment in phase 1 gain resistance during phase 2, completing the remainder of the (first line) treatment course as an infectious case with single drug resistance. This represents cases in which treatment has failed to reduce bacterial load and has allowed for the selection of resistant bacteria to dominate the infection. In addition, a small proportion (r_2) of DS cases which become non-infectious after the first phase of treatment acquire drug resistance in the second phase. This represents treatment that has cleared most of the bacterial population in the first phase, reducing both symptoms and infectiousness, but has left a pool of resistant organisms. These resistant bacteria, now given a selective growth advantage under drug treatment which has cleared competing organisms and freed resources, may now proliferate.

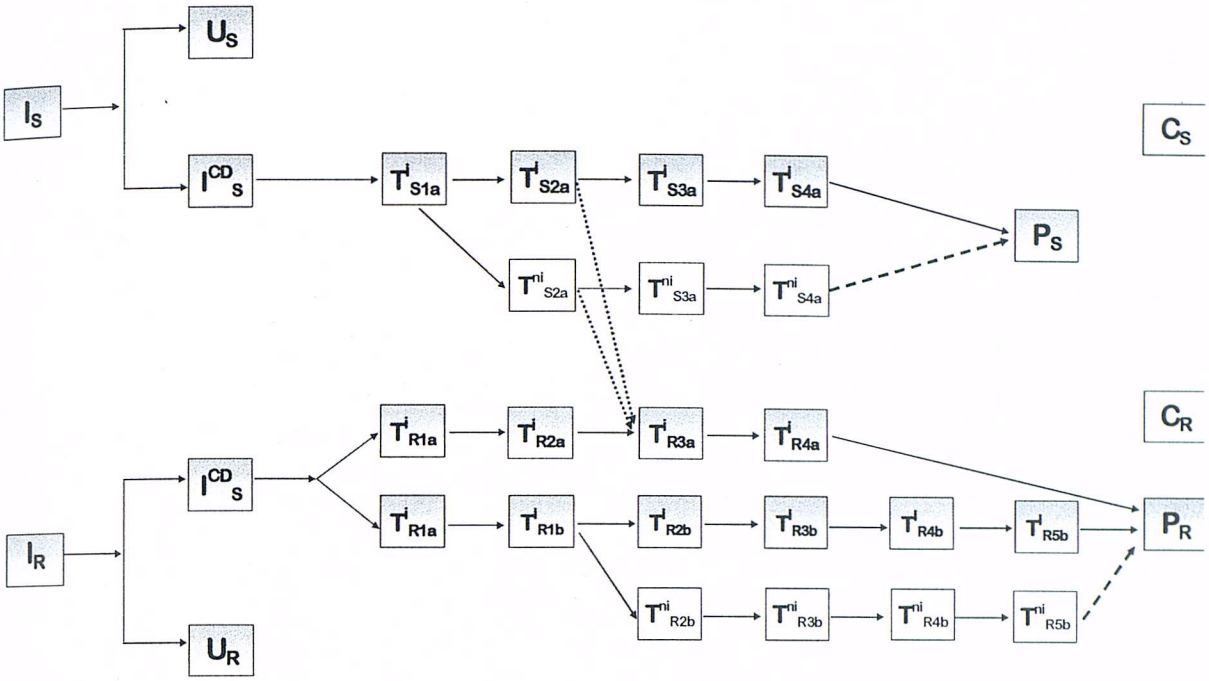


Figure 7-3. ***The model for acquired resistance in the treatment of DS cases.***

A proportion of DS cases failing treatment on regimen A acquire resistance. Treated drug sensitive cases (T_S) may be cured, fail treatment and remain drug sensitive, or fail treatment and gain resistance becoming a treated resistant case (T_R). A proportion (r_1) of treated drug sensitive cases which remain infectious in phase 2 of treatment (T^{I}_{S2a}) acquire resistance. A smaller proportion (r_2) of treated drug sensitive cases which are initially cured and become non-infectious in phase 2 of treatment (T^{ni}_{S2a}) also acquire resistance.

The first character in the subscript denotes strain type, either S or R. Darkened boxes represent infectious classes, while clear boxes represent non-infectious classes. The treated classes (T) are subdivided according to both treatment time (phases 1-4 shown in subscript), infectious status (infectious (I) or non-infectious (NI) shown in superscript), and strain type (sensitive (S) or singly resistant (R) shown in subscript). The final character in the subscript denotes the treatment regimen used: either first (a) or second line (b) treatment.

IV. E.Resistance treatment

Cases with single drug resistance undergo a similar treatment pattern. All cases are initially given first line treatment (regimen A), which is effective for drug sensitive disease. A proportion of all new incident cases, equal to pDST, receive drug testing. These test results are returned after a time (t_{DST}), and drug resistant cases are then switched to more effective treatment (regimen B for single drug resistance, regimen C

for MDR). Previously-diagnosed DR cases which have failed treatment on regimen B will, when retreated, all receive drug testing and immediately start a new course of regimen B. The repeated drug testing of these cases provides a check of the resistance profile and allows for appropriate treatment adjustments where needed.

The acquisition of multiple drug resistance (MDR) is modeled by allowing for a proportion of treated DR cases to move into a treated MDR class. The mistreatment of singly drug resistant cases with first line (regimen A) is thought to place a high selective pressure for the acquisition of higher levels of resistance. The model also provides for an increased probability of acquired MDR with time spent mistreating a DR case. This selective pressure results in a high proportion of DR cases treated with a first-line regimen developing MDR.

Cases which are initially resistant to either isoniazid or rifampicin (and are in the DR class) may develop resistance to the other drug and become an MDR case. Similarly, an initially drug sensitive case may first develop single resistance, and then MDR, passing through treated DS, DR, and MDR classes during a single course of treatment.

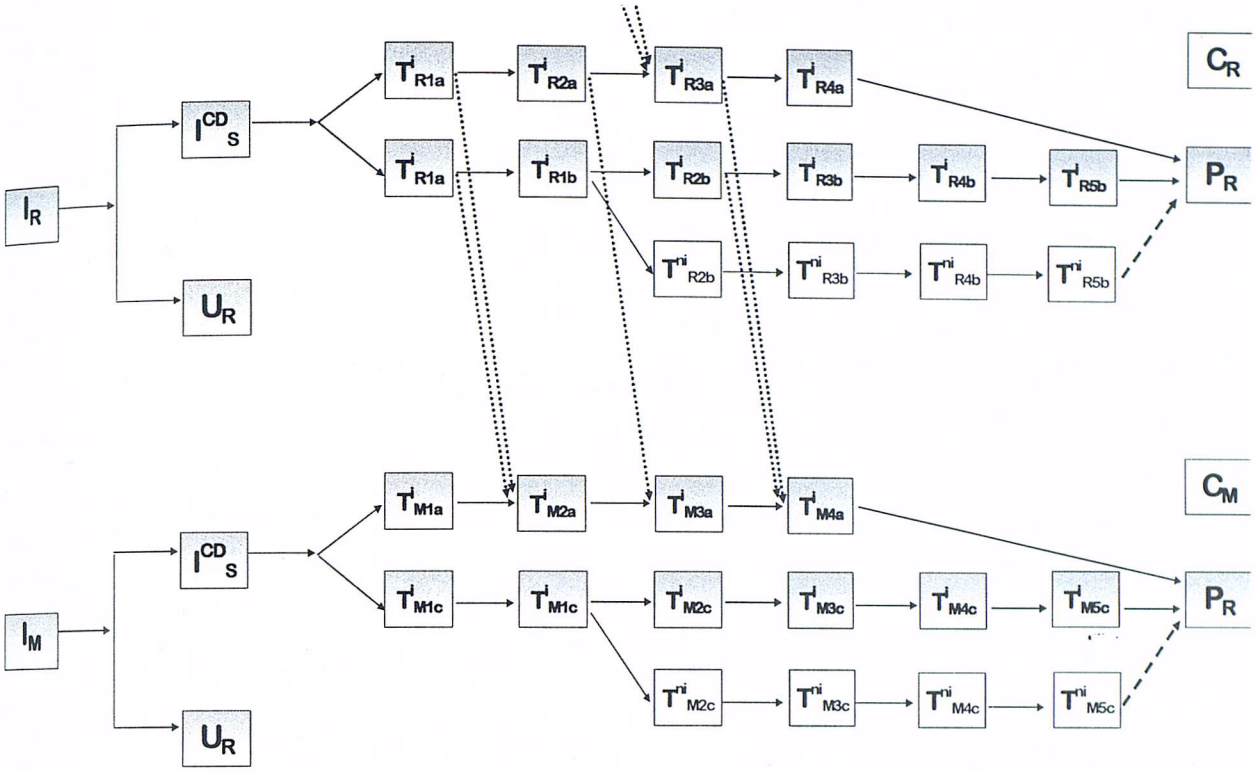


Figure 7-4. ***The model for acquired resistance in the treatment of DR cases.***

A proportion of DR cases failing treatment on regimen A or B acquire resistance. Treated singly drug resistant cases (T_R) may be cured, fail treatment and remain singly drug resistant, or fail treatment and gain higher resistance becoming a treated MDR case (T_M). R cases receiving regimen A experience a time-dependent selection for acquiring higher levels of resistance, at a rate of r_M/ month treated. In addition, a proportion (r_1) of drug resistant cases which fail treatment on regimen B remain infectious in phase 2 of treatment (T^{I}_{R2b}) acquire higher resistance.

The first character in the subscript denotes strain type, either R or M. Darkened boxes represent infectious classes, while clear boxes represent non-infectious classes. The treated classes (T) are subdivided according to both treatment time (phases 1-4 shown in subscript), infectious status (infectious (I) or non-infectious (NI) shown in superscript), and strain type (singly resistant (R) or MDR (M), shown in subscript). The final character in the subscript denotes the treatment regimen used: either regimen B for single resistance or C for MDR.

V. Model for acquired resistance during treatment regimens

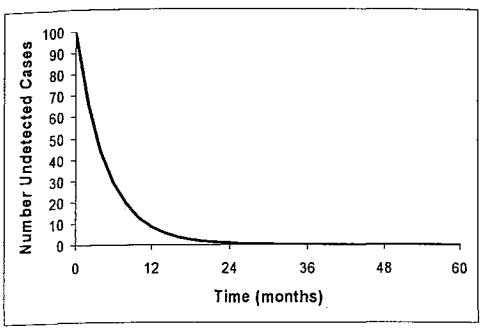
In this section, the properties and implications of this model for the acquisition of resistance during treatment are illustrated. The rates of acquired resistance depend upon the treatment regimen, strain, and infectious status. A set proportion of cases

failing an appropriate treatment regimen (first-line for DS or second-line for DR cases) acquire resistance. However, mistreated cases (first-line for DR cases) are considered to be under continuous selective pressure for resistance, such that the proportion developing resistance is positively correlated to the duration of incorrect treatment.

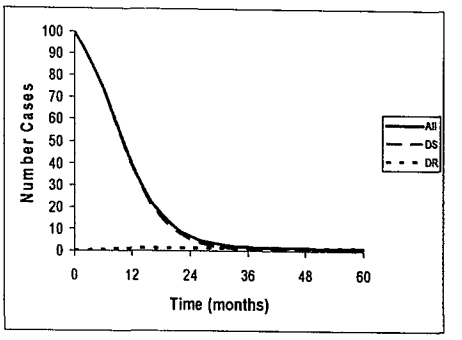
The simulations in this section follow one course of treatment for new cases of active disease. Each simulation is begun (at time 0) with 100 new infectious cases of the same strain type, either drug sensitive (DS, in simulation 1) or singly drug resistant (DR, in simulation 2). These cases are then detected, after an average time to case detection, and begin a course of treatment that lasts for 9 ½ months (for DS or DR on first-line regimen) or 12 ½ months (for DR on second-line regimen). Assuming an average time to case detection of 6 months (and a constant rate of transfer) the 100 initial cases in this simulation are all detected and have begun treatment by 20 months. (Figure 7-5A) These 100 initial cases of active disease are considered in isolation with no ongoing transmission, and are removed only through cure or death. The progressive detection and treatment of these cases (as well as self-cure and death) reduces disease until no cases remain.

Simulation 1 is begun with 100 drug sensitive cases, which progress through a single course of treatment. A proportion of these cases fail treatment, and a proportion of treatment failures acquire resistance and transfer to become singly drug resistant (DR) cases. As there is no ongoing transmission, any resistant cases present reflect acquired resistance. There is a gradual decline in prevalence of disease as DS cases are detected and receive treatment. (Figure 7-5B) Simultaneously, there is an increase in the number of DR cases, reflecting the acquisition of resistance among treated DS cases. By the end of simulation 1, 1.4% of all initial DS cases have acquired DR. (Figure 7-5C)

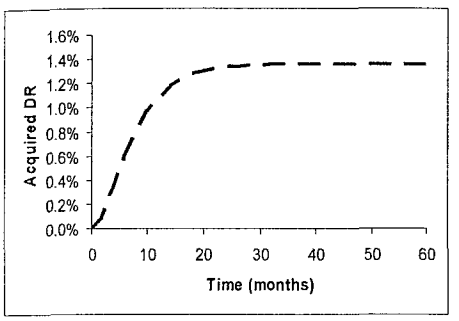
Simulation 1



5A.



5B.

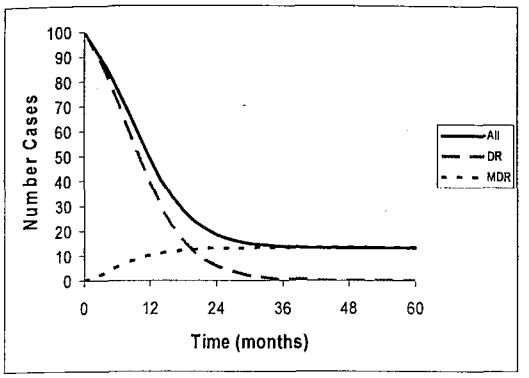


5C.

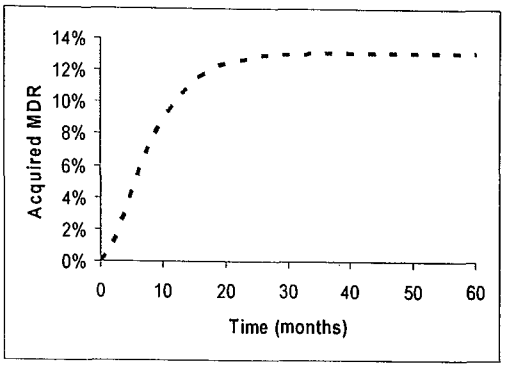
Figure 7-5. *The emergence of acquired resistance in the treatment of drug sensitive cases.* The simulation is begun with 100 infectious drug sensitive cases, which are cleared through treatment, self-cure, or death. (A) The detection and treatment of cases reduces the prevalence of drug sensitive cases, but allows for the emergence of resistant cases. (B) The number of resistant cases increases over the course of the simulation with the treatment of the drug sensitive cases. By the end of the simulation, 1.4% of the initial drug sensitive cases have acquired resistance. (C) The case detection rate is set at 80% and the treatment cure rate at 90%.

The same scenario is shown applied to the emergence of MDR from the treatment of DR cases in Figure 7-6. Note the more rapid increase in acquired MDR (than for acquired DR), reflecting the greater selective pressure assumed to exist for the mistreatment of DR cases. 14% of the initial DR cases acquire MDR during the course of treatment in simulation 2. (Figure 7-6B)

Simulation 2



6A.



6B.

Figure 7-6. ***The emergence of acquired MDR resistance in the treatment of singly drug resistant (R) cases.*** The simulation is begun with 100 infectious singly drug resistant (R) cases, which are cleared through treatment, self-cure, or death. The detection and treatment of cases reduces the prevalence of DR cases, but allows for the emergence of MDR cases. (A) The number of MDR cases increases over the course of the simulation with the treatment of the DR cases. By the end of the simulation, 1.4% of the initial DR cases have acquired MDR. (B) As in the previous simulation, the case detection rate is set to 80% and the treatment cure rate is 90%. In addition, 80% of incident cases receive drug testing and switch to a second-line regimen (with cure rate of 70%) after one month.

V. A. The emergence of acquired DR during the treatment of DS cases.

The proportion of DS cases entering treatment which acquire resistance depends upon the cure and resistance rates. During the treatment of DS cases, a set proportion of cases failing treatment will acquire DR, which includes a proportion r_1 of patients who remain infectious and r_2 of patients who become non-infectious in phase two of treatment. As either resistance rate (r_1 or r_2) is increased, so is the resulting proportion of treated DS cases which convert to DR cases. Similarly, as the cure rate for DS cases is increased, fewer cases fail treatment and fewer treated cases gain resistance. Thus, the proportion of treated DS cases which acquire DR is positively correlated to the resistance rate, and negatively correlated to the cure rate. (Figure 7-7)

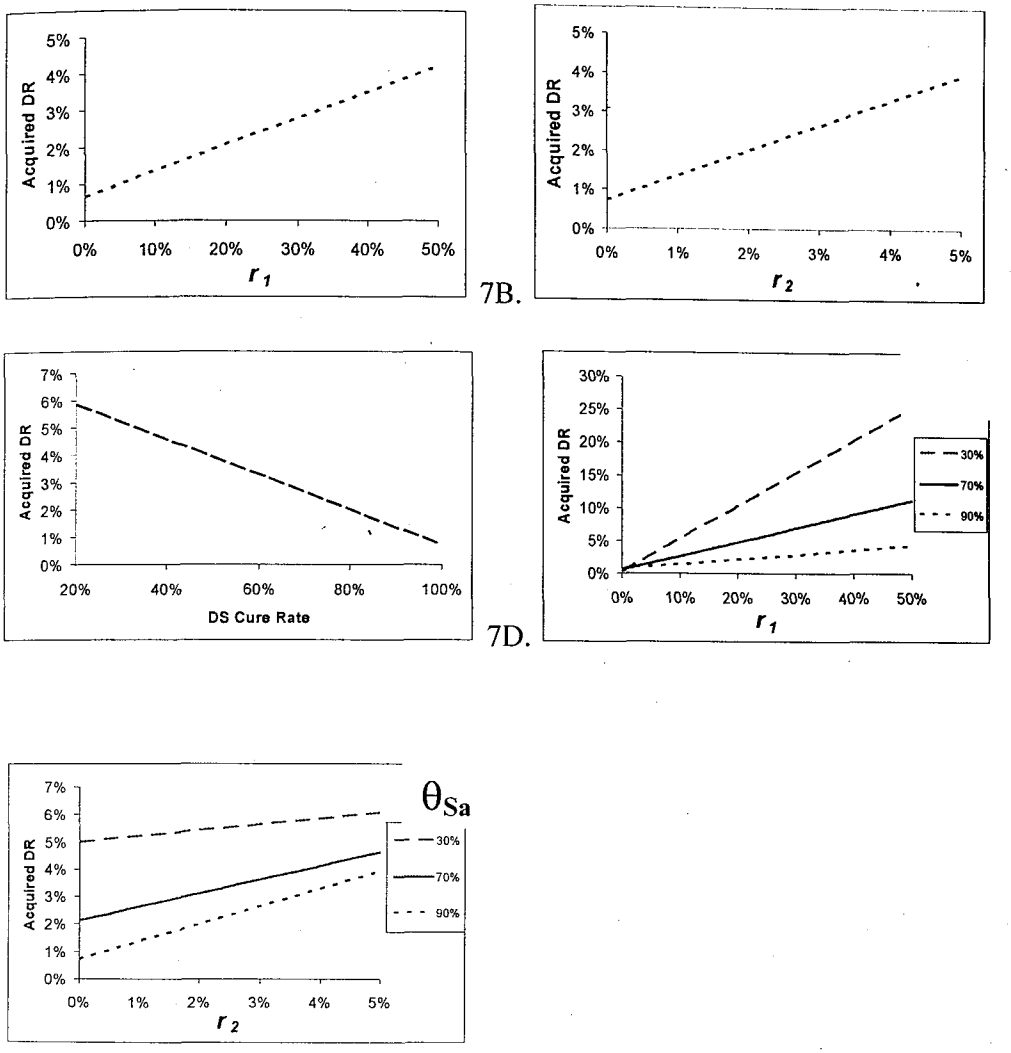


Figure 7-7. ***The rate of acquired resistance depends upon the model parameters for treatment and cure rates.*** The proportion of treated drug sensitive cases which acquire resistance is increased proportional to the probability of acquiring resistance given treatment failure (r_1 and r_2), and inversely proportional to the cure rate. In these simulations, unless otherwise indicated, control parameters are set at: Φ_{Sa} = 90%, r_1 = 10% and r_2 = 1% .

V. B. The emergence of acquired MDR from the treatment of DR cases

The proportion of all treated DR cases that acquire MDR is related to the cure rate of second-line treatment, the duration of treatment, the proportion receiving drug testing, and the time taken to return these tests. The emergence of higher levels of resistance (represented by MDR) during the treatment of DR cases is modeled at two

levels. First, DR cases mistreated with first-line regimens are assumed to experience a constant selective pressure for acquiring resistance, proportional to the treatment time. Second, a set proportion of DR cases which receive an appropriate second-line regimen, fail this treatment and develop MDR.

Drug susceptibility testing of a proportion of treated DR cases allows a switch to regimen B (with higher cure rates). Drug testing decreases the proportion of cases failing treatment, resulting in fewer cases transferring to MDR. (Figure 7-8A) The longer taken to return results of DST, the more time these cases spend mistreated with regimen A, during which there is time-dependent selective pressure to gain resistance. Thus, increasing t_{DST} results in greater numbers of acquired MDR cases. (Figure 7-8B) The rates for acquired MDR also depend upon the cure rates for DR cases, once switched to regimen B. As the DR cure rate is improved, the proportion of treated cases acquiring MDR is reduced (Figure 7-8C). At lower cure rates, more treated cases remain chronically infectious, and have a higher probability of acquiring MDR.

The proportion of treated DR cases developing MDR also depends upon the resistance rates, as defined in the model for DR cases on either regimen A or B. For DR cases on regimen A, there is a time-dependent selective pressure, r_{MDR}, defined as the proportion of cases developing MDR per month of treatment. Increasing r_{MDR} results in higher levels of acquired MDR (Figure 7-8D). For DR cases on regimen B, a set proportion of treatment failures (r_f) convert to MDR cases. As r_f is increased, so is the level of acquired MDR (Figure 7-8E). The proportion of treated DR cases which acquire MDR depends on the duration of treatment. In particular, the longer DR cases are mistreated with a first-line regimen, the greater the likelihood of developing higher levels of resistance. (Figure 7-8F)

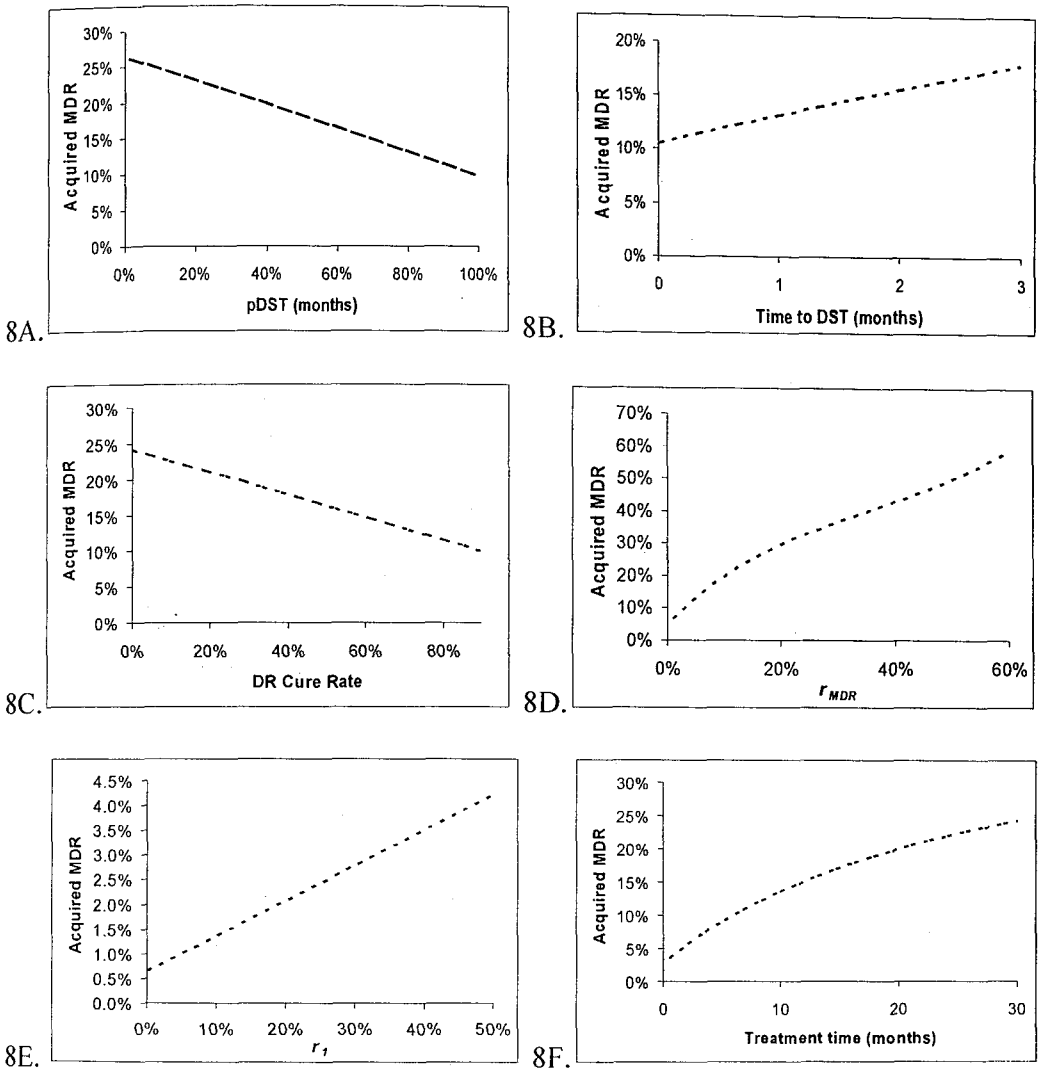


Figure 7-8. ***The rate of acquired resistance depends upon the model parameters for treatment and cure rates.*** The proportion of treated singly drug resistant (R) cases which acquire higher levels of resistance is increased proportional the probability of effective cure. Higher rates of drug susceptibility testing reduce rates of acquired MDR (A), while waiting longer for return of these drug tests allows for higher rates of acquired MDR (B). Increasing the cure rate for DR cases reduces acquired MDR (C), while increasing the probability of acquiring resistance given treatment failure (r_M and r_1) increases acquired MDR (D, E). Increasing the duration of treatment also increases acquired MDR. (F) In these simulations, unless otherwise indicated, control parameters are set at: pDST = 80%, Φ_{Rb} = 70%, t_{DST} = 1 month, r_{MDR} = 5%/month (represents the constant selective pressure during treatment of DR with regimen A), and r_1 = 10% (represents proportion of treatment failures for DR on regimen B acquiring MDR).

VI. Model for control measures

VI. A. Case detection

Surveillance programs distinguish between active and passive case detection of TB disease, with most relying on passive detection. Passive case detection refers to patients who report to health authorities with symptoms. Active case detection refers to cases found by large scale screening, which may also turn up cases without symptoms.

Passive case detection is incorporated into the model. The case detection rate is defined here as the proportion of new incident cases which are detected, diagnosed, and begin TB treatment. A proportion of new incident cases remain untreated, and this may occur for several different reasons. First, a proportion of incident infectious cases may have fewer symptoms and may therefore be less likely to seek medical attention or be tested for TB. Second, a proportion may have less access to medical services. Third, a proportion of cases may not be given appropriate and accurate diagnosis. For either of the first two possibilities, increasing the population-wide routine access to medical services should improve passive case detection, while the latter explanation would require improved proficiency in diagnosis.

VI. B. Re-treatment

The case detection rate, as defined here, does not include the (re-)detection of chronic or relapsed cases. Chronic and relapse cases are assumed to be automatically detected for re-treatment, and begin a new course of treatment after being held in a non-treated class for some time. Patients may also initially become non-infectious during the course of treatment, but later relapse to disease. Relapse is incorporated into the model by permitting a proportion of cases cured to relapse to active disease after the end of treatment.

VI. C. Case Detection and DOI

The duration of infectiousness is defined here as the time a case spends in the infectious state. The average case duration of infectiousness accounts both for those cases which are detected and receive treatment, and those which remain undetected. Chemotherapy reduces the infectious period of TB cases, by providing for more rapid cure.

Untreated cases remain infectious until they either self-cure or die. The average duration of infectiousness of an untreated case is inversely proportional to the sum of the self-cure and mortality rates ($\alpha = \mu + \mu_T + \kappa$). In this model, the average duration of infectiousness for untreated cases is approximately two years, similar to estimates in the literature.

$$\tau_{UNDET} = \frac{1}{\alpha}$$

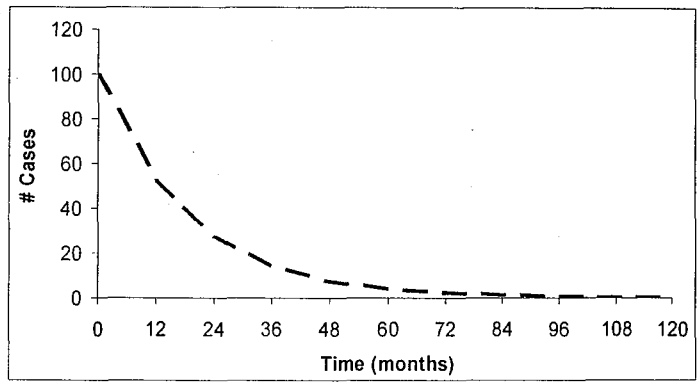


Figure 7-9. ***Timescale for removal of untreated cases.*** Untreated cases remain infectious until they either self-cure, or die from natural or TB-associated causes. 50% of cases are removed within one year, and 95% within five years. The average duration of infectiousness for these untreated cases is two years.

The duration of infectiousness of a treated case is equal to the time from case onset to the initiation of treatment (time to case detection, t_{CD}) plus the time from the start of treatment to loss of infectiousness (time spent in phase 1, t_{ph1}). The average duration of infectiousness of an effectively treated case is given in the model as:

$$\tau_{\text{treat+cure}} = \left(\frac{1}{\alpha + \frac{1}{t_{CD}}} + \frac{1}{\alpha + \frac{1}{t_{ph1}}} + \frac{1}{\alpha}\right)$$

A proportion of treated cases fail treatment and remain infectious. The average duration of treatment for these patients is given as:

$$\frac{1}{\alpha + \frac{1}{t_{CD}}} + \frac{1}{\alpha + \frac{1}{t_{ph1}}} + \frac{1}{\alpha + \frac{1}{t_{ph2}}} + \frac{1}{\alpha + \frac{1}{t_{ph3}}} + \frac{1}{\alpha + \frac{1}{t_{ph4}}} + \frac{1}{\alpha + \frac{1}{t_{ph5}}} + \frac{1}{\alpha}$$

However, as these cases remain infectious during and after the course of treatment, their average DOI is the same as that for untreated cases:

$$\tau_{treat+fail} = \frac{1}{\alpha}$$

VII. Defining duration of infectiousness by strain type

The case duration of infectiousness is derived here by strain type, assuming an epidemic of only one strain. The average duration of infectiousness for patients with either sensitive (DOI_S) or resistant (DOI_R or DOI_M) cases is given in the model by summing over the time spent in the infectious state. This is calculated as the sum of the DOI for undetected, treated and cured, and treated and not cured, and relapse cases. Each is weighted by the relative proportion of cases which follow that pathway.

VII. A. Defining the DOI for drug sensitive (DS) cases

The average duration of infectiousness for sensitive cases, as defined in the model, is equal to the sum of the following components.

Undetected

$$(1 - CDR)\ \left(\frac{1}{\alpha}\right)$$

Treated, failures

$$CDR\ (1-\phi_{Sa})\ (\frac{1}{\alpha})$$

Note: This does not include re-treatment of cases failing a treatment course. Including re-treatment would slightly reduce this average DOI, if patients started a new course in less than two years (average DOI for untreated) after infection. As the treatment course itself takes about 10 months, in addition to the time to retreatment (an average of one year), this seems a close approximation.

Treated, cured

$$CDR\ \ \phi_{Sa}\ \ (1-w_{Sa})\ \ (\frac{1}{\alpha+\frac{1}{t_{CD}}}+\frac{1}{\alpha+\frac{1}{t_{ph1}}}+\frac{1}{\alpha})$$

Note: This is the only component of the overall DOI that is reduced by treatment.

Treated, cured, relapse

$$+CDR\ \ \phi_{Sa}\ \ w_{Sa}\ \ (\frac{1}{\alpha+\frac{1}{t_{CD}}}+\frac{1}{\alpha+\frac{1}{t_{ph1}}}+\frac{1}{\alpha}+CDR\ (1-\phi_{Sa})\ (\frac{1}{\alpha})+CDR\ \ \phi_{Sa}\ \ (1-w_{Sa})\ \ (\frac{1}{\alpha+\frac{1}{t_{CD}}}+\frac{1}{\alpha+\frac{1}{t_{ph1}}}+\frac{1}{\alpha}))$$

VII. B. Defining the DOI for singly drug resistant (DR) cases

Similarly, the average DOI for cases with single drug resistance (DR) includes the following components.

Untreated

$$(1-CDR)\ (\frac{1}{\alpha})$$

Treated, no drug testing (regimen A)

$$CDR\ (1-pDST)\ (\frac{1}{\alpha})$$

Treated, drug testing, regimen B, fail

$$CDR\ \ pDST\ (1-\phi_{Rb})\ (\frac{1}{\alpha})$$

Treated, drug testing, regimen B, cured

$$CDR\ \phi_{Rb}\ pDST\ (1-w_{Rb})\ (\frac{1}{\alpha+\frac{1}{t_{CD}}}+\frac{1}{\alpha+\frac{1}{t_{DST}}}+\frac{1}{\alpha+\frac{1}{t_{ph1}}}+\frac{1}{\alpha})$$

Treated, drug testing, regimen B, cured and relapse

$$CDR\ \phi_{Rb}\ pDST\ w_{Rb}\ (\frac{1}{\alpha+\frac{1}{t_{CD}}}+\frac{1}{\alpha+\frac{1}{t_{DST}}}+\frac{1}{\alpha+\frac{1}{t_{ph1}}}+\frac{1}{\alpha})$$

VII. C. Defining the DOI for MDR cases

Untreated

$$(1-CDR)\ (\frac{1}{\alpha})$$

Treated, no drug testing (regimen A or B)

$$CDR\ (1-pDST)\ (\frac{1}{\alpha})$$

Treated, drug testing, regimen C, fail

$$CDR\ \ pDST\ (1-\phi_{Mc})\ (\frac{1}{\alpha})$$

Treated, drug testing, regimen C, cured

$$CDR\ \phi_{Rb}\ pDST\ (1-w_{Mc})\ (\frac{1}{\alpha+\frac{1}{t_{CD}}}+\frac{1}{\alpha+\frac{1}{t_{DST}}}+\frac{1}{\alpha+\frac{1}{t_{ph1}}}+\frac{1}{\alpha})$$

Treated, drug testing, regimen B, cured and relapse

$$CDR\ \phi_{Rb}\ pDST\ w_{Mc}\ \left(\frac{1}{\alpha+\frac{1}{t_{CD}}}+\frac{1}{\alpha+\frac{1}{t_{DST}}}+\frac{1}{\alpha+\frac{1}{t_{ph1}}}+\frac{1}{\alpha}\right)$$

Impact of treatment on DOI

VII. D. Higher cure rates reduce infectious period: Cure of drug sensitive cases

The duration of infectiousness is reduced by effective treatment. As more cases are detected and given treatment, the average infectious period is shortened. This depends on the effectiveness of treatment, represented in the model by the cure rate (θ). Thus, the average DOI for a DS case depends both on the proportion of cases receiving treatment (CDR) and on the cure rate for drug sensitive cases (with regimen 'A', θ_{Sa}). When the DS cure rate θ_{Sa} is set to 0%, treatment does not lead to faster cure, and the average DOI for DS cases remains the same as for untreated cases. As the cure rate θ_{Sa} is increased (shown ranging from 20-100%), more cases are cured at a faster rate, and the DOI-DS is reduced in proportion to the level of case detection. (Figure 7-10)

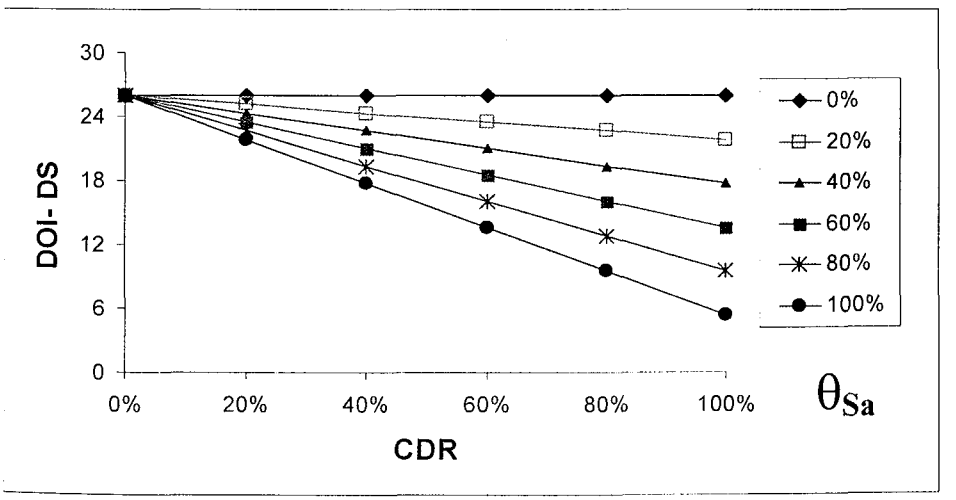


Figure 7-10. ***Effective treatment reduces the duration of infectiousness of drug sensitive cases.*** The DOI of DS cases is reduced by higher levels of case detection and treatment (CDR). The cure rate for DS cases on regimen A (θ_{Sa}) is varied from 0% to 100%. When the cure rate is set to 0%, there is no effective cure, and the regardless of treatment rate, the DOI remains the same as for untreated cases. Higher

cure rates allow for effective cure and reduce the DOI of DS cases in proportion to the CDR. The average case duration of infectiousness (DOI) for drug sensitive cases is shown in months.

VII. E. Cure of drug resistant cases

The DOI for resistant cases is similarly reduced by increasing levels of case detection and treatment efficacy. Shown below is the average duration of infectiousness for MDR cases, over a range of case detection (CDR) and treatment cure (treatment of MDR cases on regimen 'C', θ_{Mc}) rates. When treatment with regimen 'C' is completely ineffective at curing MDR cases (θ_{Mc}= 0%), then the DOI-MDR is the same as for untreated cases. As the θ_{Mc} cure rate is increased, case detection and treatment reduces the DOI of MDR cases. (Figure 7-11)

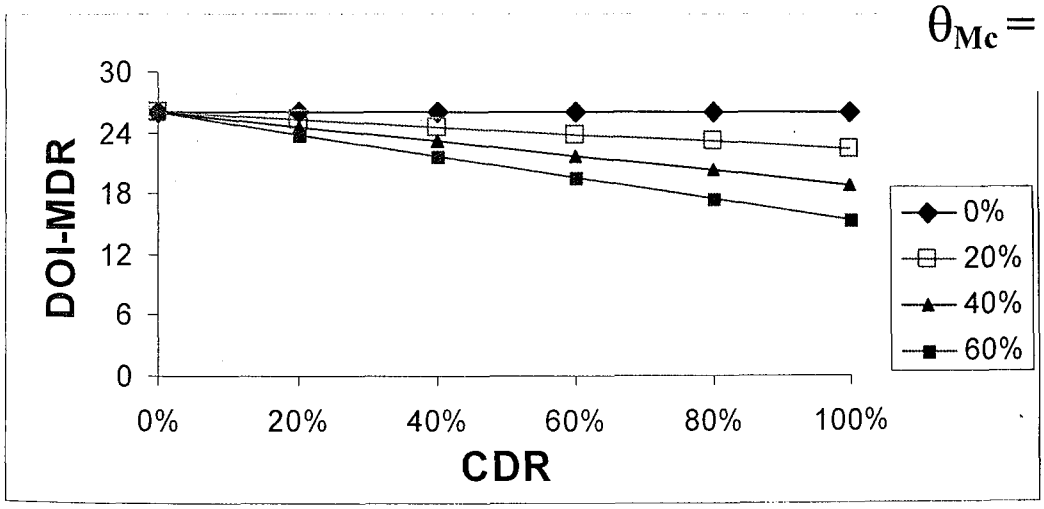


Figure 7-11. ***Effective treatment reduces the duration of infectiousness of MDR cases.*** The DOI of MDR cases is reduced by higher levels of case detection and treatment (CDR). The cure rate for MDR cases on regimen C (θ_{Mc}) is varied from 0% to 60%. Higher rates of treatment and cure reduce MDR case DOI. The proportion of incident cases receiving drug susceptibility testing (pDST) is set to 90%. The average case duration of infectiousness (DOI) for MDR cases is shown in months.

VII. F. Higher levels of drug testing increase effective treatment, reduce infectious period

The average case DOI for resistant cases (DR or MDR) are also reduced by increasing levels of drug susceptibility testing. As a higher proportion of incident cases are given drug testing and shifted to regimens with higher treatment efficacy, more resistant cases are cured, and the DOI is shortened. Shown below is the DOI for MDR cases, under varying levels of treatment (CDR, 0% - 100%) as well as proportion of treated cases receiving drug testing (DST, 0%, 30%, 60%, 90%). For a given treatment rate increasing the DST rate results in further decreases the DOI, as cases are given effective regimens that allow for faster cure. (Figure 7-12)

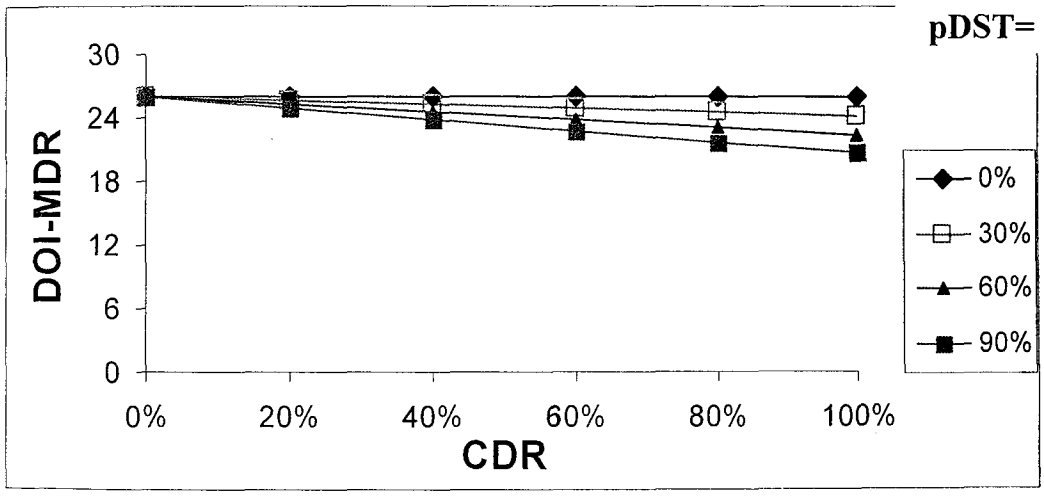


Figure 7-12. ***Drug susceptibility testing (DST) reduces the duration of infectiousness of MDR cases.*** When there is no drug testing (pDST = 0%), the MDR case DOI is equal to that of untreated cases, as all incident MDR cases are given first line treatment (regimen 'A' for drug sensitive disease). As a higher proportion of newly detected cases are given drug testing (range 30-90% shown), the MDR-DOI is reduced. With higher levels of case detection and higher levels of drug testing, the average MDR-DOI is reduced. An MDR cure rate on regimen 'C' (θ_{Mc}) is set to 50%. The average case duration of infectiousness (DOI) for MDR cases is shown in months.

VII. G. Comparing strain-specific case DOIs

The case duration of infectiousness is reduced in proportion to the extent of effective treatment. For drug sensitive cases this is a function of case detection and cure rates, while for resistant cases, the proportion receiving drug testing must also be included. Treatment reduces the case DOI according to the proportion effectively cured—highest here for DS cases, followed by DR and then MDR cases. (Figure 7-13)

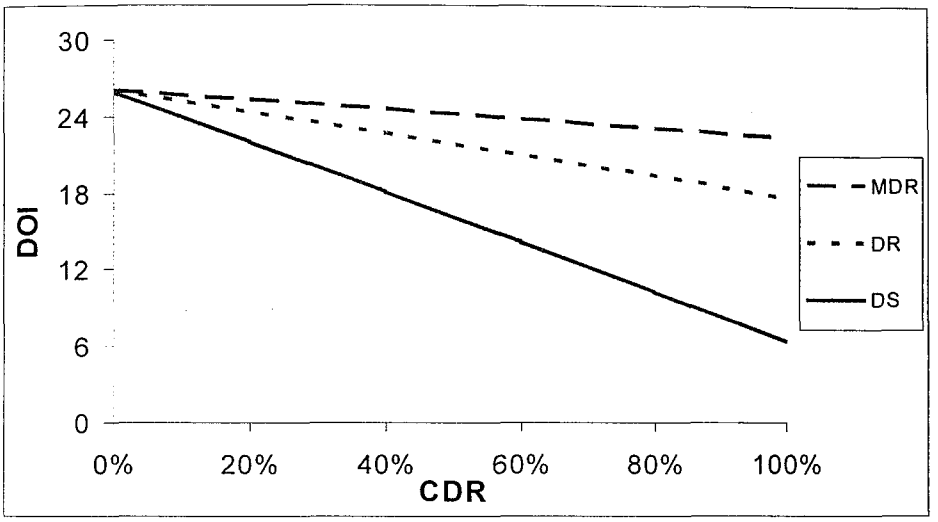


Figure 7-13. ***The average DOI for DS, DR and MDR cases.*** **The detection and treatment of cases reduces the average duration of infectiousness.** For a given treatment rate (CDR), the DOI of sensitive cases are reduced to a greater extent than that for either DR or MDR cases. The proportion of new incident cases given drug susceptibility testing (pDST) is set to 90%. The cure rates for DR on regimen B (θ_{Rb}) = 70%, and for MDR on regimen B (θ_{Mc}) = 30%. For DS cases, the reduction in DOI by level of treatment depends on θ_S. For resistant cases, the reduction in DOI with case detection depends on the proportion receiving DST, allowing for switch to a second line regimen, as well as the cure rates (θ_{Rb} for DR and θ_{Mc} for MDR). The average case duration of infectiousness (DOI) for sensitive (DS), singly resistant (DR), and MDR (MDR) cases is shown in months.

VIII. Parameter Table

Table 7-1. ***Parameter Table.***

CONTROL PARAMETERS		
Symbol	**Name**	**Explanation**
CDR	Case detection rate	Proportion of incident cases receiving treatment
tCD	Time to case detection and treatment	Time from case onset to detection for treatment
ΦSa	TcSa	Treatment cure rate for DS cases on regimen A
ΦRa	TcRa	Treatment cure rate for DR cases on regimen A
ΦRb	TcRb	Treatment cure rate for DR cases on regimen B
ΦMa	TcMa	Treatment cure rate for MDR cases on regimen A
ΦMc	TcMc	Treatment cure rate for MDR cases on regimen C
w1	Relsa	Relapse rate for DS cases on regimen A
r1	Acquired resistance rate	Acquired resistance rate for infectious DS case under regimen A- or infectious DR case under regimen B
r2	Acquired resistance rate	Acquired resistance rate for non-infectious DS case under regimen A
rMDR	Acquired resistance rate	Acquired resistance rate for infectious DR case under regimen A
w2	Relrb	Relapse rate for DR cases on regimen B
pDST	DST rate	Proportion of treated cases receiving drug susceptibility testing
tDST	Time to DST	Time taken from start of treatment to return drug susceptibility tests and start second line treatment
tph1..4	Time each treatment phase	Phase 1: 2 weeks Phase 2: 3 months Phase 3: 3 months Phase 4: 3 months
tph5	Time to re-treatment	1 year

STATE VARIABLES

Symbol	Explanation	Strain type	Infectious?
X	Susceptible		-
L	Latent	LS - LR- LM	-
NI	Non-infectious disease		-
I	New infectious cases	IS - IR- IM	+
U	Untreated infectious cases	US -UR-UM	+
T	Treated cases	TS -TR-TM	+ / -
P	Previously failed treatment	PS - PR- PM	+
C	Cured	CS - CR- CM	-

TREATMENT STATE VARIABLES				
Symbol	Description	Strain type	Infect-ious	Treatment regimen
T_{Sla}	Sensitive infectious cases receiving treatment a	S	+	a
T_{Sa}	Sensitive non-infectious cases receiving treatment a	S	-	a
T_{Rla}	Resistant infectious cases receiving treatment a	R	+	a
T_{Rlb}	Resistant infectious cases receiving treatment b	R	+	b
T_{Rb}	Resistant non-infectious cases receiving treatment b	R	-	b
T_{Mla}	MDR infectious cases receiving treatment a (or b)	M	+	a or b
T_{Mlc}	MDR infectious cases receiving treatment c	M	+	c
T_{Mlc}	MDR non-infectious cases receiving treatment c	M	-	c

IX. Discussion

IX. A. The costs of poor treatment

The benefit of treatment, in this model, comes from the reduction in case duration of infectiousness. This in turn reduces ongoing transmission and disease levels. If the sole effect of treatment is to decrease case DOI, then higher treatment

rates would always lead to reductions in disease, as shown in the simulations above. While this may generally be true, several possible exceptions are considered below.

i. Poor treatment and selection for resistance

First, treatment selects for drug resistance. As more cases receive treatment, the rates of drug resistance often increase. A number of studies have suggested correlations between poor treatment and acquired resistance. This includes errors in patient treatment including delayed diagnosis, use of an inappropriate regimen such as adding one new drug to a failing regimen (effective monotherapy), and irregular dosing due to limited drug supply. In addition, patient factors including treatment default and poor adherence increase rates of resistance. These factors each reduce the overall treatment cure rates and may increase the risk of developing resistance. (Iseman 1999)(Mahmoudi, Jama, 93)(Chaulet and Zidouni 1998)

In the model developed here, this relationship is modeled by allowing a proportion of treatment failures to acquire drug resistance. Thus, increasing the treatment cure rates reduces acquired resistance. Because of this assumed association between treatment failure and acquired resistance, maintaining low cure rates in a population will select for resistance. Further, maintaining these low cure rates while expanding treatment coverage may increase the level of acquired resistance.

ii. The effect of case detection on average case duration of infectiousness

In the absence of second line treatment, drug resistant cases experience the same average duration of infectiousness as untreated cases. Accordingly, as the resistance prevalence grows within a population under a first line control program, the average case DOI will approach that of untreated cases. Consequently, under some circumstances adding higher levels of treatment may lead to increases in the average

case DOI. This occurs in our model when there is no available second line treatment for resistant cases, high case detection, low first line cure rates, and a high probability of acquiring resistance. When there is high resistance prevalence, increasing detection rates when only first line treatment is available may increase the average case DOI. This would increase transmission and exacerbate the resistance problem.

iii. Poor treatment may increase patient's duration of infectiousness?

Finally, it is may be possible that poor treatment can increase an individual patient's duration of infectiousness. Early studies noticed that shortly after introducing treatment into a population, case numbers increased. Many of these cases were chronically infectious and non-responsive to treatment, but not all appeared to have resistance. sSome researchers have suggested that, independent of resistance status, poor treatment may increase a patient's duration of infectiousness beyond that of untreated cases. The first noticeable impact of treatment is reduction in mortality, allowing infectious cases to live longer. It is more difficult to achieve successful cure, so under suboptimal treatment regimens, mortality may be reduced with out much increase in cure rates. This could result in selection for chronic infectious cases, whose lives are prolonged under this suboptimal treatment that does not fully cure cases. (Johnston R.F. 1974; Gangardharam and Jenkins 1997; Mitchison 1997)

Possible explanations for prolonged duration of infectiousness can be found at a mycobacterial level, and include selection for persistent mycobacteria. Suboptimal treatment regimens may select for semi-dormant bacteria, which would be cleared by stronger treatment, but under weak selection are able to survive and replicate at a slow rate. This would leave the host still symptomatic, but perhaps with lower infectiousness, bacterial load and mortality rates. This explanation is consistent with studies that have demonstrated lower rates of transmission from chronically infectious

cases after failing multiple courses of treatment. (Lutwick 1995; Davies 1998; Iseman 2000)

If the above phenomenon does occur, then treatment with low cure rates would incur an additional cost of increasing case duration of infectiousness. The implications of this are serious in planning control of epidemics. If the cost of poor treatment is significant enough, it is possible that there would be circumstances in which it would be better to work on improving the implementation of the program, including improvements to treatment completion and reducing relapse rates, before considering expansion of coverage to untreated patients. This would also be a reason to favour implementation of DOTS programs, which work to ensure standardized regimens and high rates of treatment completion. Further experimentation is necessary to explore the relationship between the range of possible aspects contributing to poor treatment, and the outcome in terms of duration of infectiousness, mortality and cure rates, and resistance.

X. Summary of Findings

In this chapter, the model for the emergence and transmission of TB was expanded to include additional aspects of control and treatment, including case detection, treatment times, multiple regimens, and drug susceptibility testing. The impact of treatment on duration of infectiousness was examined in terms of strain type, treatment regimen and efficacy. Higher rates of treatment and cure were shown to reduce the DOI. For drug sensitive cases the case DOI was shown to be inversely correlated to the treatment and cure rate. For drug resistant cases, the case DOI is also inversely correlated to the drug susceptibility testing rate.

The dynamics and timescale for emergence of acquired resistance during treatment in a population were explored within the model formulation. The proportion

of treated drug sensitive cases acquiring resistance is positively correlated to the treatment rate and the probability of acquiring resistance given treatment failure, and inversely correlated to the cure rate. For the acquisition of MDR during the treatment of DR cases, the rate is also inversely correlated to the drug testing rate, and positively correlated to the time taken to return drug tests and the duration of treatment.

In the next two chapters, this model is used to consider the impact of various control strategies on the course of TB epidemics, and to formulate priorities for control in different settings.

8. Chapter 8

Control of resistant tuberculosis

8. CHAPTER 8

CONTROL OF RESISTANT TUBERCULOSIS

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III. RESULTS: SIMULATIONS FOR FIRST LINE CONTROL PROGRAMS

III. A. SIMULATIONS FOR FIRST LINE CONTROL PROGRAMS
III. B. ENDEMIC SETTING
III. C. TREATMENT BENEFITS: BALANCE TREATMENT AND CURE
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V. A. TOP PRIORITIES IN TB CONTROL:
V. B. NEXT PHASE IN TB CONTROL
V. C. TREATMENT AND SELECTION FOR RESISTANCE: STRIKING A BALANCE
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V. F. DRUG RESISTANCE

VI. SUMMARY OF FINDINGS

I. Goals

I. A. To examine the impact of elements of first line TB control programs on the course of epidemics, including notification and cure rates.

I. B. To examine the importance of treatment waiting times within a control program.

I. C. To examine the impact of drug testing and second line resistance treatment on the course of an epidemic.

II. Abstract

Control strategies for TB epidemics are explored using the mathematical models defined in Chapter 7. Case detection is shown to be the most important control measure required for reduction of disease. A strong first line control program, which includes high case detection and cure rates, leads to rapid reduction of disease. However, treatment may select for resistance, particularly if low cure rates have been maintained while increasing treatment coverage. In order to reduce resistance prevalence and prevent further resistant epidemics, a second line control program that includes drug susceptibility testing and specialized treatment regimens is required. These second line control measures rely on a strong first line program, and should be implemented only once a high proportion of all cases are detected and treated successfully.

In previous chapters, dynamics of resistance epidemics, including the emergence and transmission of resistance have been explored. In Chapter 7, the model was expanded to include additional aspects for control and treatment. In this chapter, the model is used to investigate the effects of different TB control strategies.

III. Results: Simulations for first line control programs

III. A. Simulations for first line control programs

In this section, simulations are presented to illustrate the impact of first line control measures. In each simulation, an epidemic is begun by the introduction of one infectious case into a susceptible population, and run until an equilibrium level of disease is reached. In this simulation, the epidemic was first allowed to progress for 100 years in the absence of any treatment, allowing disease levels to reach equilibrium

with an incidence of 110 cases per 100,000 and prevalence of 240 cases per 100,000. (Figure 8-1) In the following simulations shown in Figures 8-2 though 8-5, a first line control program is introduced after the epidemic has reached equilibrium. These simulations vary both by epidemic state (disease burden) and level of control (case detection and treatment).

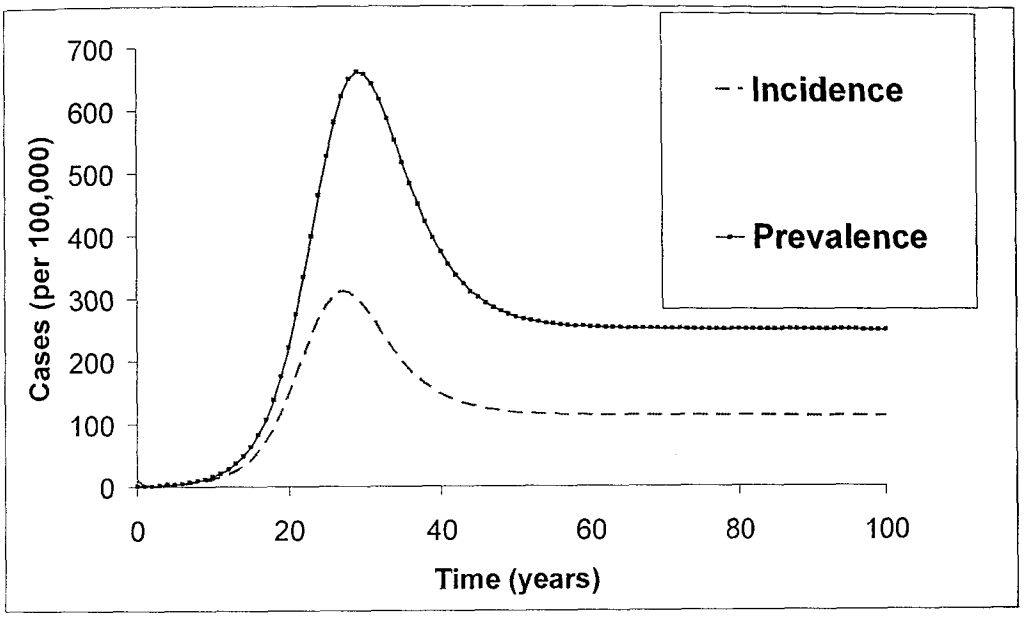


Figure 8-1. ***The impact of treatment on a young epidemic.*** Simulations were begun by introducing one infectious case into a susceptible population of size 10^8, and allowing an epidemic to emerge in the absence of treatment. The incidence rate (given as new cases per 100,000) and prevalence (as cases per 100,000) are followed over the course of the simulations.

III. B. Endemic setting

In first simulations examine the effect of introducing treatment into an endemic setting, in which disease levels have stabilized at an equilibrium level. Shown in Figure 8-2 is an example of an epidemic that progressed for 100 years in the absence of any treatment, allowing disease levels to reach equilibrium with an incidence of 110 cases per 100,000. Introducing any curative treatment will disrupt this equilibrium, reducing the average case duration of infectiousness and transmission. Even low treatment rates, such as case detection of 30% (with a cure rate of 85%), reduces

incidence and prevalence of disease. Higher treatment rates results in a faster and greater decline in incidence and prevalence. (Figure 8-2)

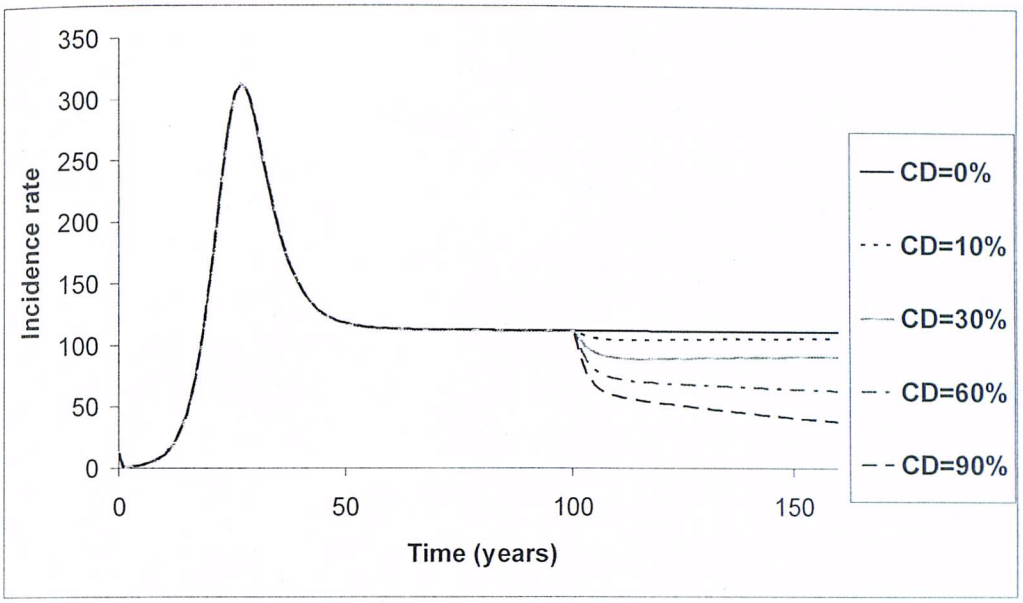


Figure 8-2. ***Impact of introducing treatment into a high burden endemic setting.*** Treatment is introduced into an epidemic at equilibrium levels with a high burden of disease. Simulations were first run to allow the epidemic to reach equilibrium in the absence of treatment. After 100 years, disease reached equilibrium with an incidence of 120 cases per 100,000. Treatment was then introduced, with a case detection rate (CDR) of 0%, 10%, 30%, 60% or 90% of incident cases. This control program was maintained for 60 years. When no treatment is introduced (CDR=0%), incidence rate remains at equilibrium levels. As higher treatment rates are introduced, there is a reduction in both incidence and prevalence of disease over the course of the epidemic.

The relationship between treatment and disease levels, in the context of this high burden endemic setting, is summarized in Figure 8-3. At higher treatment rates, the disease levels are reduced over the course of the control phase of the simulations. Shown is the final incidence by the treatment level (the case detection rate) used during the control phase. Higher treatment rates result in a greater and faster decrease in disease. (Figure 8-3)

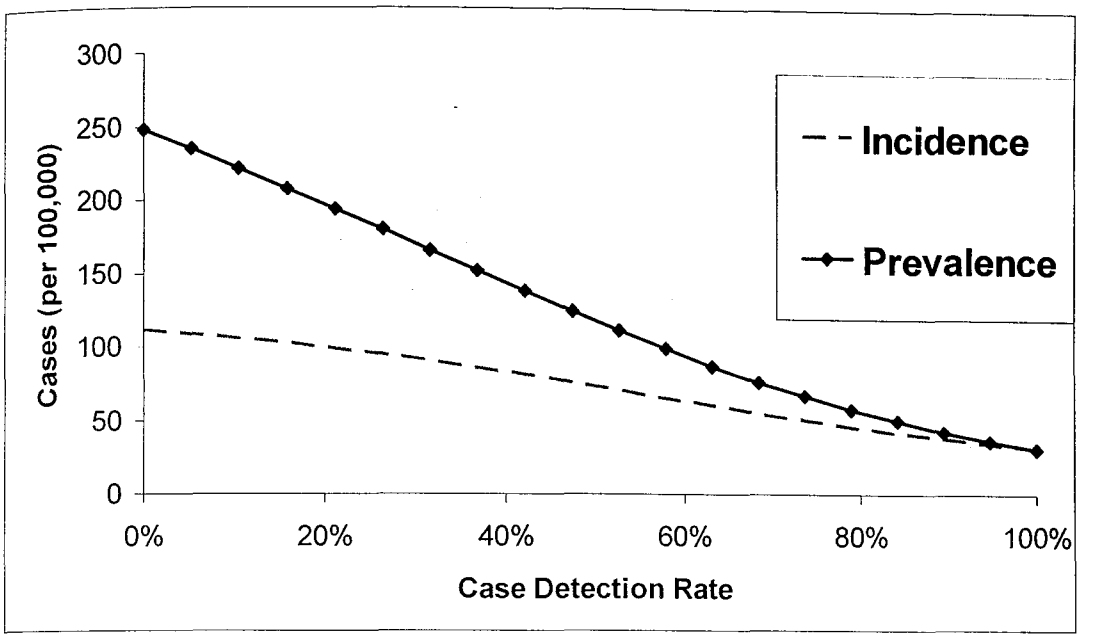


Figure 8-3. ***Introducing treatment into an endemic setting reduces disease.*** Higher treatment rates result in lower incidence and prevalence of disease. Simulations were run as in Figure 8-2. During the control phase of the simulation, the case detection rate was set at values ranging from 0% to 100%. The final incidence of disease is shown as the new infectious cases per 100,000 after 100 years of treatment is shown. Treatment rates are shown as the proportion of incident cases receiving treatment (Case Detection Rate).

III. C. Treatment benefits: balance treatment and cure

Treatment reduces disease prevalence by reducing case duration of infectiousness and transmission. Successful reduction of disease requires not only detection, but also effective treatment and cure. However, there is a balance between case detection and treatment cure. In terms of disease prevalence, the major burden is in detecting cases—even with only a mildly effective cure, there will be a reduction in disease prevalence. Conversely, increasing the cure rates for all treated patients will only have a significant impact on the course of the epidemic if a large proportion of incident cases are detected.

Even with low cure rates, wide scale treatment is beneficial in reducing disease burden. The impact of varying treatment cure rates is examined in a setting of a high incidence epidemic at equilibrium, with an incidence rate of 100 cases per 100,000. After the epidemic reached equilibrium levels of disease, treatment was introduced with

a given case detection rate, and treatment cure rate set at 20, 60, or 90%. Even at the low cure rate of 20%, there is a significant decrease in disease. At the higher cure rates of 60% or 90%, there is a faster and greater reduction in disease levels. (Figure 8-4) This illustrates that, even at low cure rates, it is beneficial to detect and treat as many cases as possible.

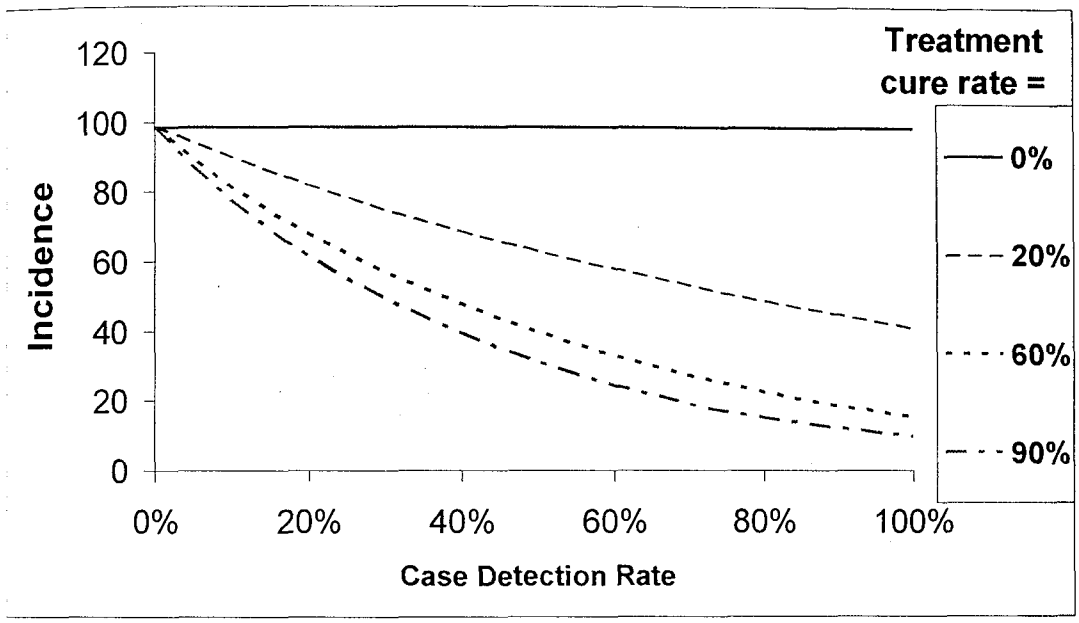


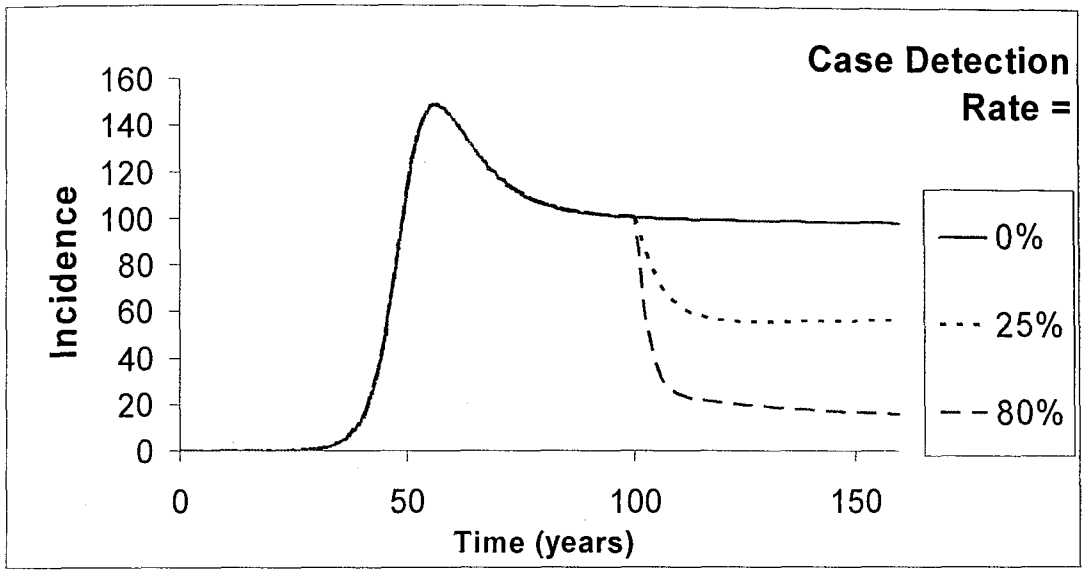
Figure 8-4. ***Balance between treatment and cure rates: reduction of disease.*** Introducing treatment with any of the given cure rates results in a notable decrease in incidence and prevalence of disease. More disease is prevented at higher case detection rates. In addition, at higher detection rates, the treatment cure rate has a more significant impact on disease prevented. As more cases receive treatment, improvements to the treatment efficacy result in a greater reduction in disease.

These simulations were run as in Figure 8-2. During the 60 years of control, the case detection rate was set at a value between 0% and 100%, and the treatment cure rate was set at 20%, 60%, or 90%. The final incidence rate is shown in (C) as new cases per 100,000.

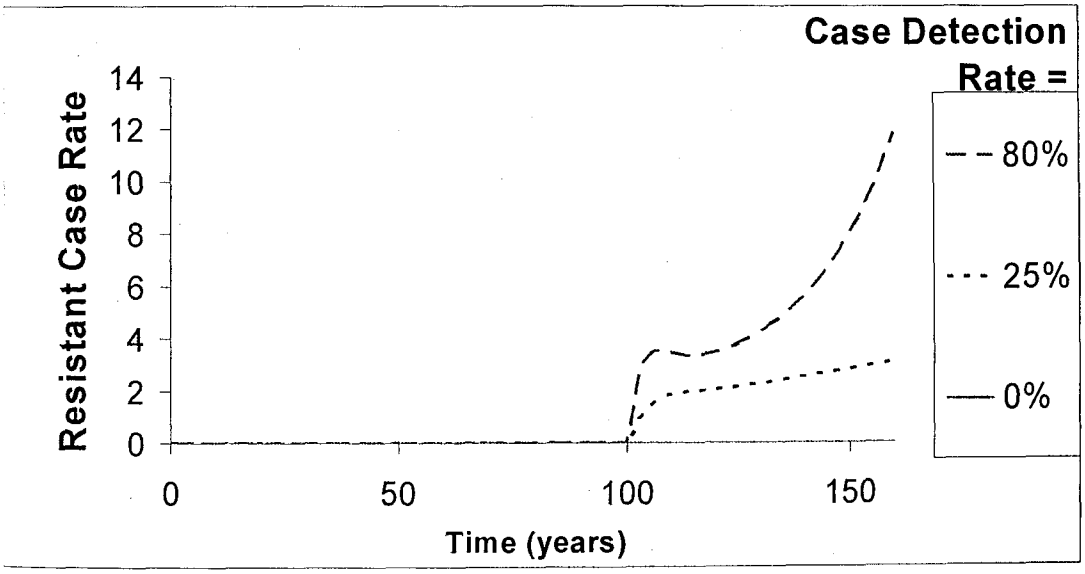
III. D. Selection for resistance under poor treatment and high detection

The relationship between low cure rates and resistance is examined in the context of a high burden setting. Simulations were first run to equilibrium in the absence of treatment, reaching stable disease levels with an incidence of 100 cases per 100,000. Treatment was then introduced, reducing incidence while selecting for

resistance. (Figure 8-5A,B) The highest levels of resistance are seen under high detection and low cure. (Figure 8-6) Thus, increasing the case detection rates without simultaneously increasing cure rates can promote higher prevalence of resistance and possibly disease.



7A.



7B.

Figure 8-5. ***Selection for resistance under high treatment.***

Higher levels of case detection reduce disease incidence (A), while increasing resistance prevalence (B). Simulations were run as in Figure 8-2. During the control phase of the simulation, treatment was begun with a case detection rate of 0%, 25%, or 80%. The treatment cure rate was set at 90%. The incidence rate is shown as new cases per 100,000 (A) and the resistant case rate as cases per 100,000 (B).

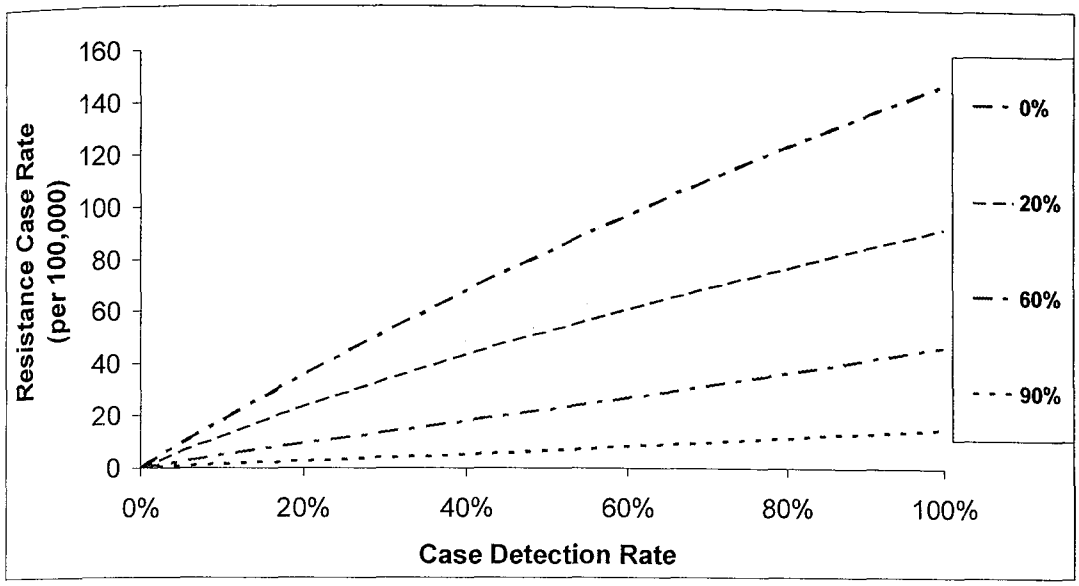


Figure 8-6. ***Selection for resistance under high treatment and low cure rates.*** The greatest selection for resistance occurs under high rates of treatment combined with low cure rates. The prevalence of drug resistant cases increases with case detection, and declines with higher treatment cure rates. At higher rates of case detection, changes in the cure rate have a greater impact on resistance prevalence. The highest resistance prevalence occurred under high case detection combined with low cure rates. Simulations were run as in Figure 8-2. During the 60 years of control, the case detection rate was set at a value between 0% and 100%, and the treatment cure rate at 20%, 60%, or 90%. The resistant case rate is shown as the number of resistant cases per 100,000 population.

III. E. Time to case detection (waiting time to treatment)

The time to treatment, as defined the model, represents the time taken from disease onset (and case infectiousness) until the start of treatment. As explained in the methods section, this includes the time taken for a new case to enter a health system, receive correct diagnosis, and begin tuberculosis treatment. The first depends upon onset of patient symptoms, access to medical resources, and willingness to report. The second depends upon the passive screening program, including familiarity with recognizing symptoms of tuberculosis and using appropriate diagnostic tests, availability of these diagnostic tests, and the time taken to return these tests. The third depends upon availability and access to appropriate drug regimens as well as administrative programs.

III. E. 1. Waiting times and case DOI

The longer patients wait until beginning treatment, the longer their duration of infectiousness. Even high rates of treatment and cure will have limited effect in reducing case DOI if cases spend long waiting for treatment. The shortest waiting time shown in the following simulations is one month. This represents rapid reporting, allowing for just a minimum estimated time for patients to recognize symptoms, report to a health service, receive diagnostic tests, confirm diagnosis, and begin treatment. In our model, an average waiting time of 1 month, combined with a case detection rate of 80% and treatment cure rate of 90%, the case duration of infectiousness is 8 months. As the waiting time is increased to 4 months, the DOI is increased to 10 months. With a waiting time of 12 months, the DOI is increased to 13 months. In comparison, the average case DOI in the absence of any treatment is 24 months. (Figure 8-7)

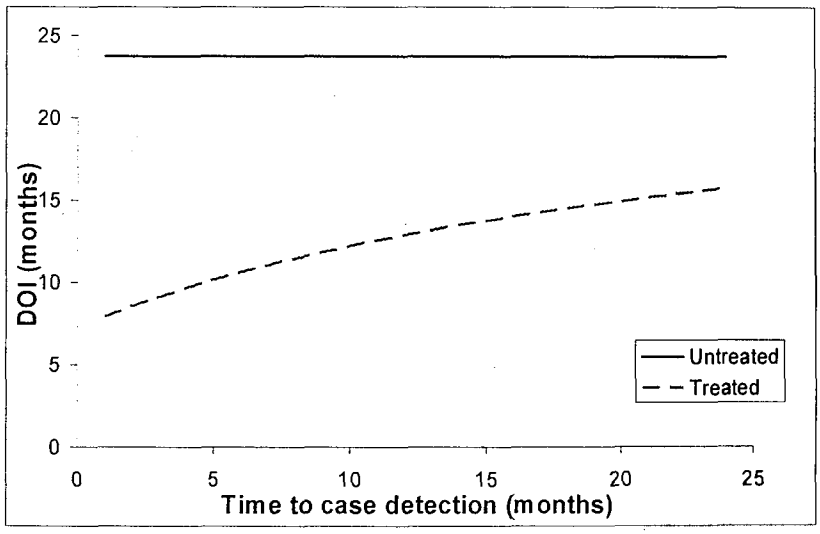


Figure 8-7. *Longer treatment waiting times result in increased average duration of infectiousness of treated cases.* Shown is the average case duration of infectiousness (in months) of treated and untreated cases. Simulations were run as in Figure 8-2. As the average waiting time to treatment is increased from 1 to 24 months, the average duration of infectiousness of treated cases is increased from 7 to 16 months. The average duration of infectiousness of untreated cases remains at 24 months.

III. F. Waiting times and disease levels

We examine how treatment waiting times affect the course of an epidemic in a high burden setting. In these simulations, the epidemic is run for 100 years with no treatment, reaching an equilibrium level of disease with an incidence rate of 100 cases per 100,000 and disease prevalence of 0.8%. Then a strong first line treatment program, with case detection rate of 80% and treatment cure rate of 90%, is introduced and maintained for the next 60 years. While the detection and cure rates are held constant, the average waiting time between case infectiousness and treatment is varied between simulations. (Figure 8-8) As the treatment waiting time is increased from one month to either four or twelve months, the incidence and prevalence of disease over the course of the epidemic are increased. (Figure 8-8)

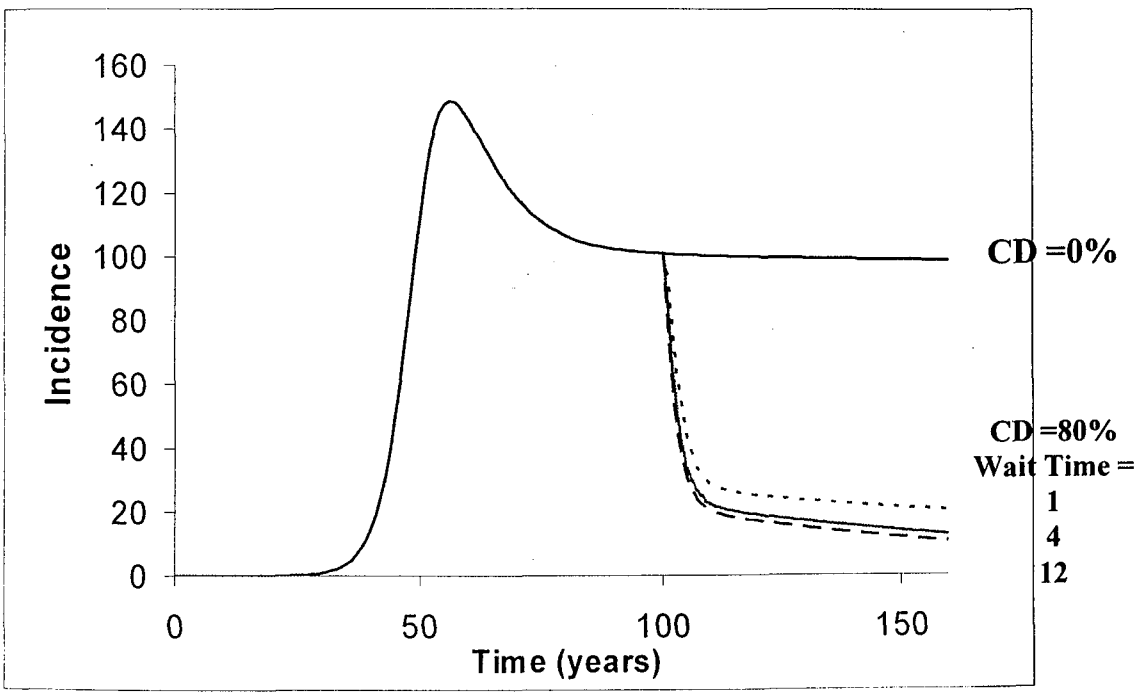


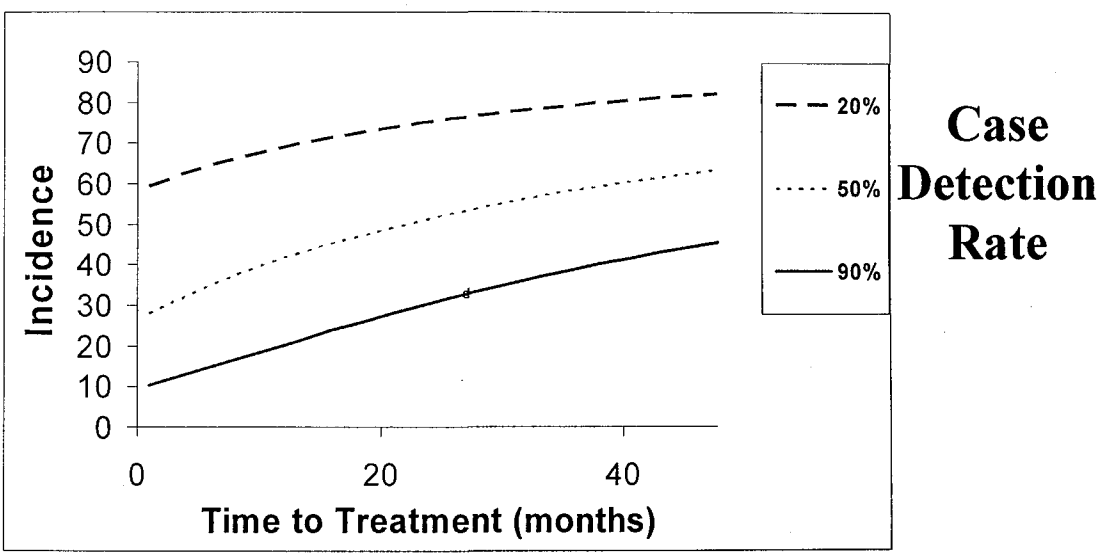
Figure 8-8. ***Decreasing the waiting time to treatment reduces incidence of disease.*** For the same detection and cure rates, shortening the waiting time to treatment resulted in reduced incidence of disease over the course of the epidemic. As a control, a simulation is shown in which no treatment is introduced (CDR = 0%), and the disease levels remain at equilibrium. Simulations were run as in Figure 8-2. A first line control program was then introduced after 100 years, with a case detection rate of 80% of incident cases and a treatment cure rate of 90%. The average case waiting time to treatment was set at 1, 4, or 12 months. This control program was maintained for 60 years.

In Figure 8-9, simulations of the same epidemic are shown, in which the waiting times are varied from 1 to 48 months. Longer waiting times allow for increased duration of infectiousness and transmission, and result in increased incidence and prevalence of disease. (

Figure 8-9A) In addition, at longer waiting times, there is less treatment and selection for acquired resistance. As expected, increasing the case waiting time slightly reduces resistance. This effect is more significant at higher levels of case detection, as higher treatment rates provide additional selection for resistance. (

Figure 8-9B)

A.



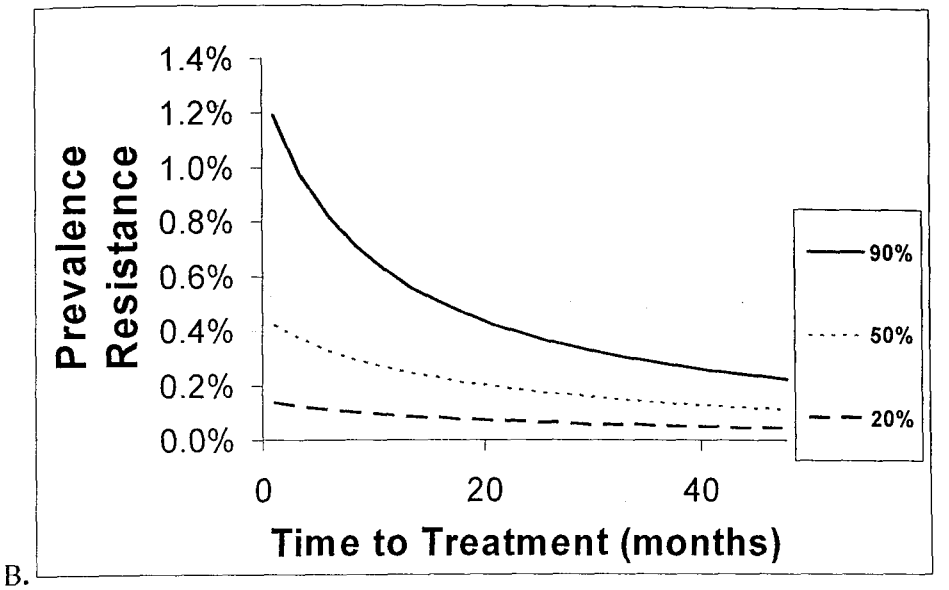


Figure 8-9. ***Waiting time to treatment: increases disease and reduces selection for resistance.*** For a given rate of case detection, increasing the waiting time to treatment increases incidence of disease (A) while resulting in lower selection for resistance (B). This effect is more pronounced at higher levels of case detection.

Simulations were run as in Figure 8-8. During the treatment phase of the simulation, the case detection rate was set at 20%, 50%, or 90%, and the average case waiting time (Time to Case Detection) was set at values between 1 and 48 months. The final incidence of disease (A, new cases per 1000,000) or resistance prevalence (B, the proportion cases resistant at end of simulation) after 60 years of constant treatment with the given level of detection, is shown against the time to treatment.

IV. Simulations for second line control programs

IV. A. Simulations

In this section, the impact of second line control for resistance is explored. Epidemics were simulated under varying level of control, which includes case detection, drug testing, and first and second line treatment. These simulations were run in three phases. In the first phase, one infectious drug sensitive case is introduced into a susceptible population, and allowed to spread in the absence of treatment for 100 years, reaching equilibrium with an incidence rate of 100 cases per 100,000. In the second phase of the simulation, a strong first line control program is introduced, with a case detection rate of 80% and treatment cure rate (for drug sensitive cases) of 90%, and maintained for 30 years. In the third phase of the simulation, the first line control

program is supplemented with second line, including drug testing and treatment of resistant cases.

IV. B. Second line control effects course of epidemic

The first line control reduces incidence (Figure 8-10A), but allows for the selection of resistance (Figure 8-10B). When no second line control is introduced (pDST=0%, Figure 8-10), the prevalence of resistance continues to increase. In comparison, when the first line program is supplemented with drug testing and resistance treatment of 40% or 90% of incident cases (pDST= 40%, 90%, Figure 8-10), there are further reductions in disease incidence over the course of the epidemic, and lower resistance prevalence. (Figure 8-10)

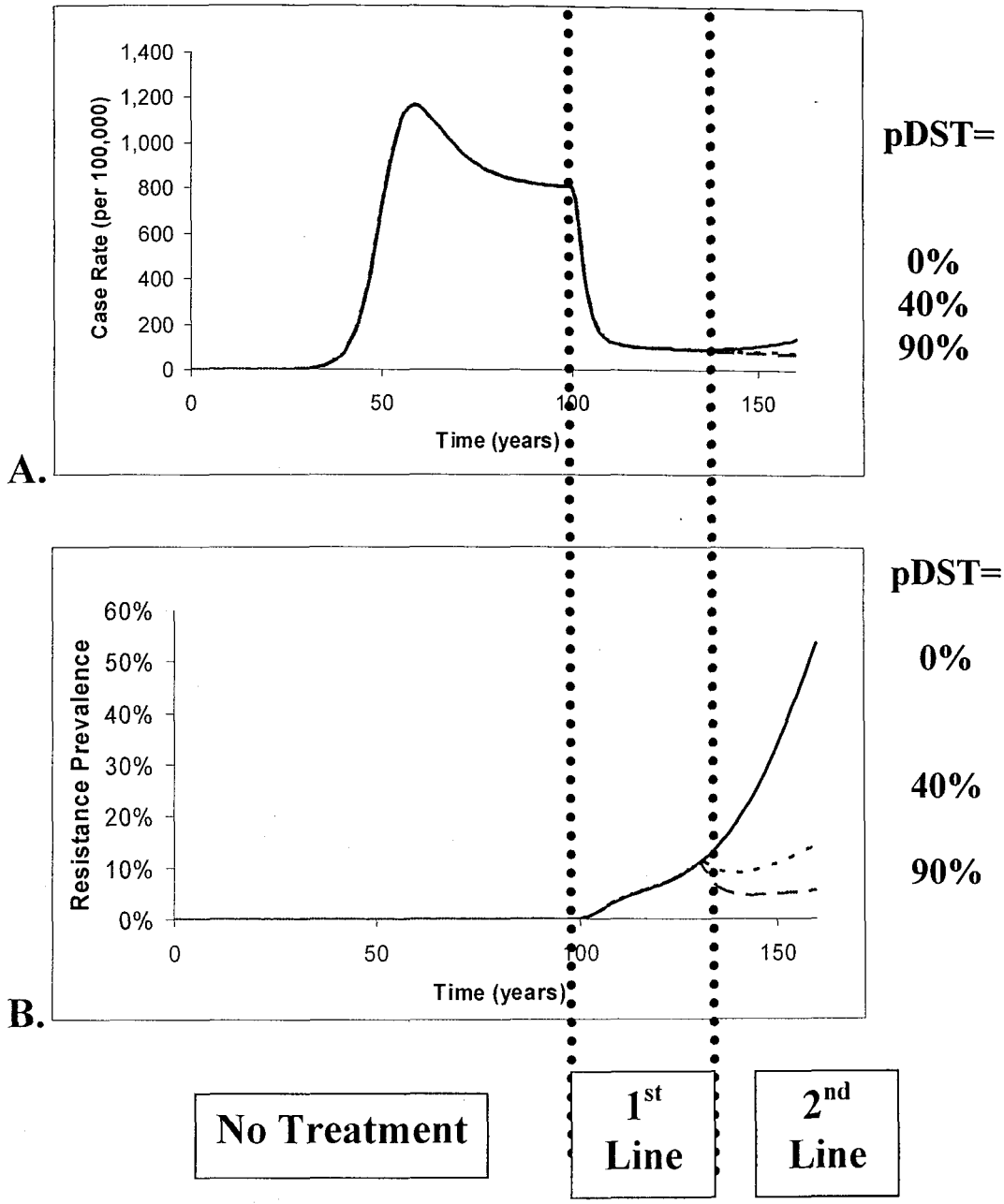


Figure 8-10. ***Introducing second line treatment reduces disease and resistance over course of epidemic.***

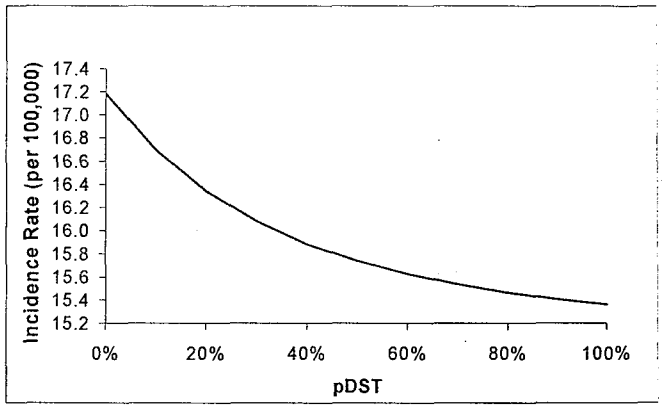
During the first phase of the simulation, the epidemic was run for 100 years under no treatment and reached an equilibrium level of disease with an incidence rate of 100 cases per 100,000. During the second phase of the simulation, lasting for thirty years, first line control was added with a case detection rate of 80% and a drug sensitive cure rate of 90%. The third and final phase of the simulation lasted for an additional thirty years. During this final phase second line treatment was introduced in addition to the first line control. The prevalence of disease is shown in (A) as the number of cases per 100,000 population, and the resistance prevalence in (B) as the proportion of cases which are resistant.

IV. C. Proportion cases receiving drug testing

All cases receiving treatment begin with a first line regimen. A proportion of treated cases (p_{DST}) receive drug susceptibility testing. Only after drug testing results have been returned and show resistance are cases shifted to a second line regimen. Drug testing is assumed to be accurate and allows for the correct diagnosis of drug resistance patterns with a 100% positive predictive value. Sensitivity of DST is not specified as distinct parameters in the model. Rather, the parameter for the DST rate reflects the overall "true rate" of detection of resistant cases. This includes the use of drug susceptibility tests, and the accuracy and sensitivity of detection.

The drug testing of new incident cases reduces the prevalence of both disease and resistance. Higher levels of drug testing reduce resistance, and to a lesser extent, disease prevalence. (Figure 8-11B)

A.



B.

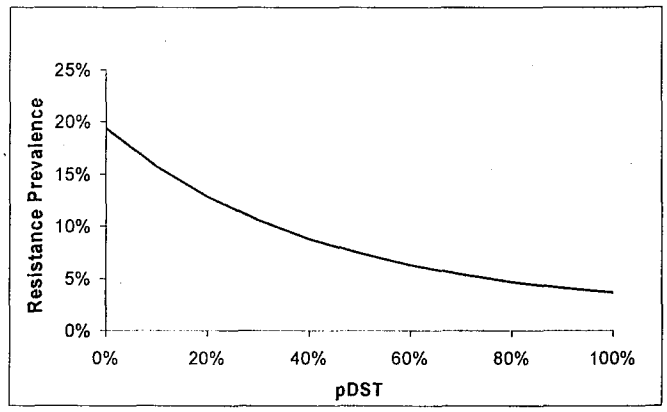


Figure 8-11. ***Drug testing and resistance treatment reduce disease and resistance.*** At higher rates of drug testing, both the incidence of disease and resistance prevalence was reduced. The rate of drug testing is given as the proportion of incident cases receiving drug susceptibility testing and appropriate treatment (pDST).

Simulations were run as in Figure 8-10. During the third phase of the simulation, the proportion of cases receiving drug testing (pDST) was varied from 0%-100%. Shown is the incidence rate of new cases per 100,000 (C) and the resistance prevalence (the proportion of cases are resistant) (D) at the end of the simulation.

IV. D. Second line treatment efficacy

The implementation of drug testing programs can only be useful when combined with an effective second line cure for resistant cases. In figure 8, the impact of introducing second line control, with a range of resistance cure rates, is examined. These simulations were first run to equilibrium levels of disease in the absence of treatment. Then a strong first line control program was introduced with a case detection rate of 80% and cure rate for sensitive cases of 90%. After 30 years of this first line control, a second line program was added, with varying levels of drug testing and resistance cure rates. As shown in figure 8A, during the first treatment phase (only first line treatment), the prevalence of resistance rapidly increased. The addition of second line treatment in the final phase of the simulation resulted in a reduced resistance prevalence, compared with that under no second line treatment.

The treatment cure rates for sensitive and singly resistant cases were set fixed at 90% and 70%, respectively. The cure rate for multiply resistant cases was set at 50%, 25%, or 0%. When the cure rate for MDR cases is assumed to be completely ineffective (cure=0%), drug testing and treatment of resistant cases still results in a reduction in resistance. This reflects the detection and treatment of singly resistant cases. This in turn reduces the emergence of MDR. As the cure rate for MDR cases is increased to 25% or 50%, the detection of resistant cases results in further reduction in resistance. (Figure 8-12)

This shows that even with low MDR cure rates, the second line control program results in a decrease in resistance prevalence. Drug testing and resistance treatment is useful even if can treat only a proportion of resistant cases. This treatment reduces the pool of general resistance. This in turn reduces the emergence of MDR. In addition, even low MDR cure rates are higher than that under first line treatment, and thus reduce case DOI and ongoing resistance transmission.

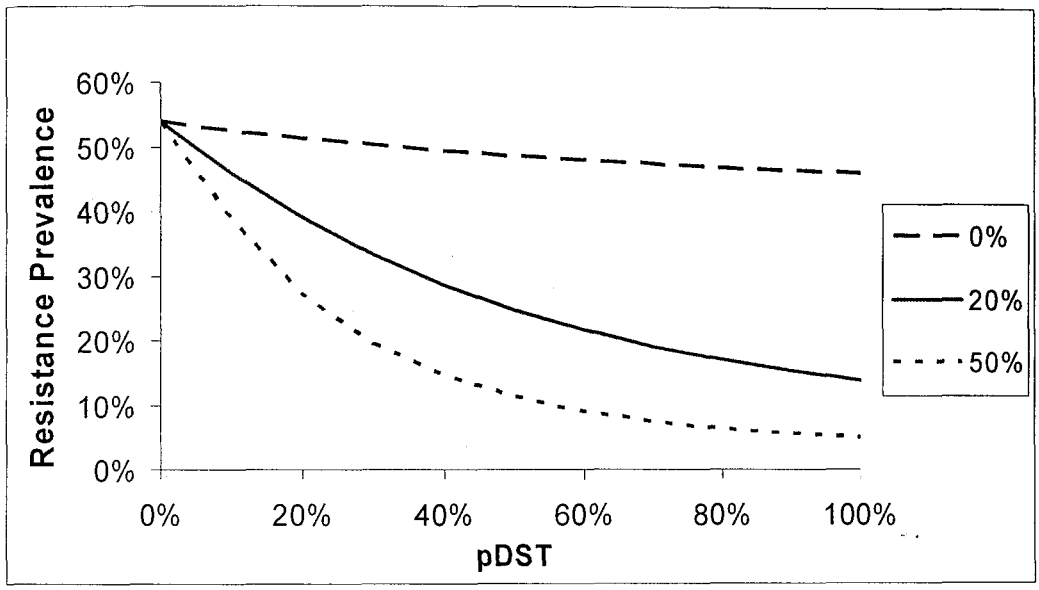


Figure 8-12. ***The impact of drug testing varies by efficacy of treatment for multiply resistant cases.*** At higher treatment cure rates for multiply resistant cases, the implementation of drug testing results in lower resistance prevalence. Even at low cure rates for multiply resistant cases, there is some reduction in resistance with greater drug testing.

Simulations were run as in Figure 8-10. During the first phase of the simulation, the epidemic was run for 100 years under no treatment and reached an equilibrium level of disease with an incidence rate of 100 cases per 100,000. During the second phase of the simulation, lasting for thirty years, first line control was added with a case detection rate of 80% and a drug sensitive cure rate of 90%. The third phase, lasting for an additional thirty years, included drug testing of a proportion of incident cases (pDST) varied from 0% to 100%. The treatment cure rate for sensitive cases was set at 90%, for singly resistant cases at 70%, and for multiply resistant cases to 0%, 20%, or 50%. Shown is the final resistance prevalence (the proportion of cases resistant) at the end of the simulation.

IV. E. Built on strong foundations

The impact of drug testing also depends on the first line control program. In particular, cases must be detected before receiving drug testing and second line treatment. In the simulations shown in Figure 8-13, second line treatment is introduced

into epidemics under varying levels of first line control. At higher levels of case detection, the same level of drug testing will result in a greater reduction in resistance. At low levels of case detection, implementing drug testing of even a large proportion of incident cases may not stop an emerging resistant epidemic. At these low levels of case detection, the more effective means to control the epidemic and the emergence of resistance would be to increase detection and treatment rates. (Figure 8-13) In these simulations, the lowest level of resistance is found under low case detection. Conversely, the most resistance occurs under high case detection, which provides more opportunity for resistance to emerge. Once resistant epidemics have emerged, higher rates of drug testing and second line treatment are required to reduce resistance prevalence. (Figure 8-13)

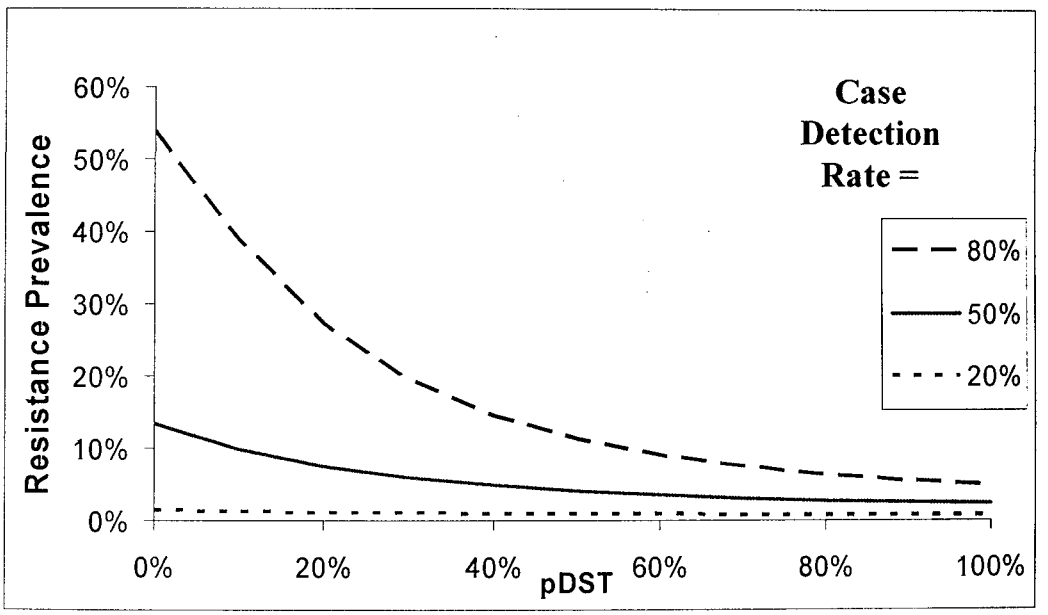


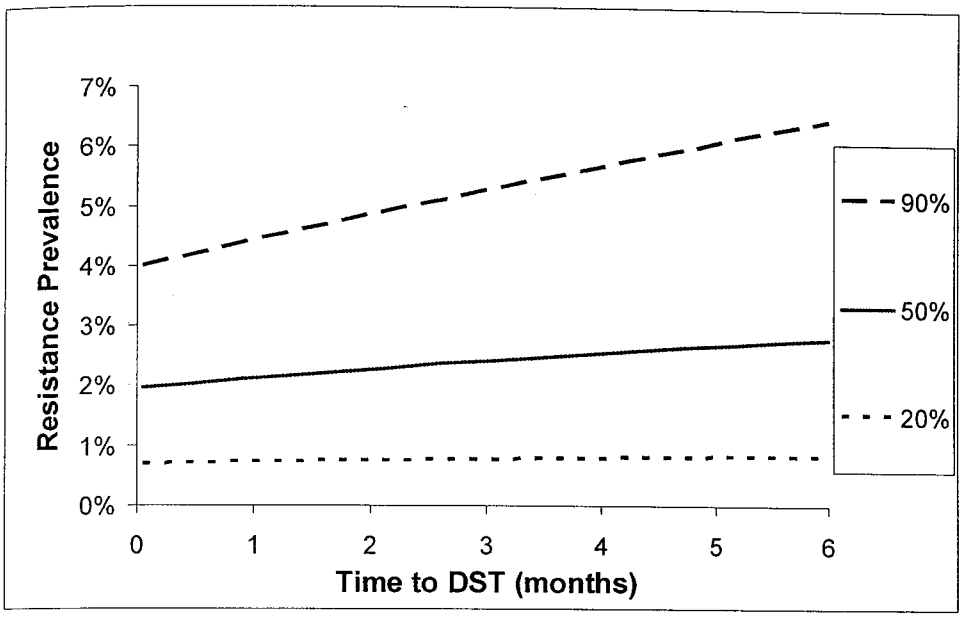
Figure 8-13. ***Impact of drug testing by case detection rate.*** At higher rates of case detection, the implementation of drug testing results in greater reduction in resistance prevalence.

Simulations were run as in Figure 8-10. During the second and third phases of the simulations, the case detection rate was set at 20%, 50%, or 80%. During the third phase of the simulation, the proportion of incident cases receiving drug testing (pDST) was varied from 0% to 100%. The treatment cure rate for sensitive cases was set at 90%, for singly resistant cases at 70%, and for multiply resistant cases at 50%. Shown is the final resistance prevalence (the proportion of cases resistant) at the end of the simulation.

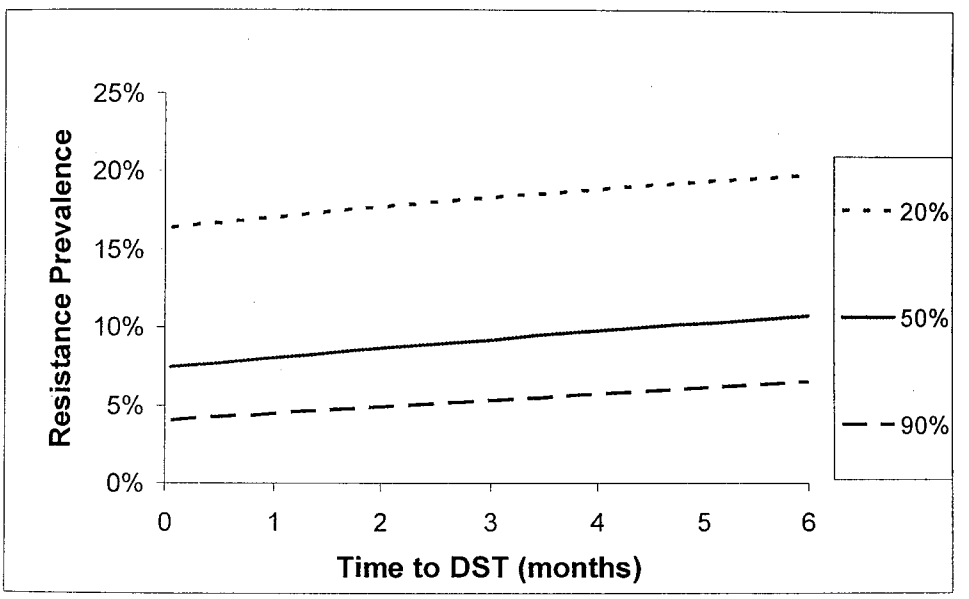
High rates of case detection may select for resistance – particularly if combined with low first line cure rates. However, high case detection also provides the opportunity to find and treat resistant cases. Thus, introducing drug testing on top of high case detection offers a chance to make significant reductions in resistance. In comparison, under low case detection it may not be possible to treat a high proportion of resistance cases, but lower selection for resistance may mean there is less of a resistance problem to repair.

IV. F. Time to return drug testing results: waiting time to resistance treatment

The time taken to return drug testing results has a limited impact on the level of resistance. Longer waiting times to return drug testing results and begin resistance treatment results in slight increases in resistance prevalence. The impact of reducing the waiting time for drug tests is greatest in situations with high case detection (Figure 8-14A) and resistance (Figure 8-14B). At higher rates of case detection, the same reductions in waiting times results in a greater decrease in resistance prevalence. (Figure 8-14)



A.



B.

Figure 8-14. ***Time to return drug susceptibility testing results.*** Shown is the final resistance prevalence (the proportion of cases resistant) at the end of the simulation. At longer waiting times to switch to appropriate second line regimens, there is an increase in resistance prevalence.

Simulations were run as in Figure 8-10. During the third phase of the simulations, the waiting time to return drug susceptibility tests and switch to an appropriate second line regimen was set to values between 1 and 12 months. During the third phase of the simulation, the proportion of incident cases receiving drug testing (pDST) was set at 0%, 50%, or 90%. The case detection rate during the second and third phases was set at 90%. The treatment cure rate for sensitive cases was set at 90%, for singly resistant cases at 70%, and for multiply resistant cases at 50%.

V. Discussion

V. A. Top priorities in TB control:

In this chapter, priorities in control of TB epidemics are explored. The first and most important control measure for reducing disease burden is ensuring a high level of case detection. Second, maintaining high treatment cure rates will reduce disease as well as limit the emergence of resistance. Introducing a control program with high detection and cure rates will rapidly reduce the disease levels in a population. The main findings in this chapter are summarized as follows:

1. Case detection is the central priority.

2. There is a balance between treatment and cure—in terms of reducing disease burden, higher case detection is a priority. For prevention of resistance, ensuring high cure rates is most important, and treatment coverage should not be expanded until target cure rates are met.

3. A strong first line control program, with high case detection and cure rates, will rapidly reduce disease levels.

4. Additional control measures, on top of a strong first line control program, will further reduce disease and prevent resistance.

These findings are supportive of current WHO recommendations for global TB control. The WHO's primary priorities in global TB control are increasing global case detection and treatment cure rates, and to this end, have set targets to achieve global detection of 70% of cases with treatment success rate of 85%. These studies also reaffirm that the most important first steps in control are establishing the effective treatment of a high proportion of cases in the population. Only after a strong control program with high levels of case detection and treatment success is established, should further control measures be considered. (WHO 2003; WHO 2003)

V. B. Next phase in TB control

Once this strong first line control has been implemented, and disease burden has been reduced, supplementary control measures may be considered. Additional control measures will allow for further reductions in disease, as well as treatment and prevention of resistance. These programs can be geared towards the next stage in TB elimination.

The other control measures considered in this chapter include:

1. reducing treatment waiting times
2. drug susceptibility testing
3. specialized treatment of resistance cases

Each of these control measures were shown to be beneficial in situations where there was already high case detection and cure rates. Reducing treatment waiting times results in further reduction in disease. However, it should be noted that this is small in comparison to the reduction in disease caused from increased levels of case detection and cure. The benefits of reducing the average case waiting times are highest under control that includes high detection and cure, as this leads to the greatest reduction in average case duration of infectiousness.

Introducing these control measures results in reduction of disease, and for the latter two, reduction in resistance. Each of these measures provides the most benefit when introduced into a control program that includes high detection and cure of drug sensitive cases. Further, each of these measures provides only small benefits, in terms of reduction in disease, relative to increases in case detection and first line treatment cure rates. These control measures should be considered as additions to already strong first line control programs, in low incidence regions where there are sufficient resources for expansion of control.

V. C. Treatment and selection for resistance: striking a balance

The balance between case detection and cure rates is measured in terms of both disease and resistance. When considering the benefit of treatment in preventing disease, the most important concern is increasing coverage. The higher the proportion of incident cases receiving treatment, the greater the reduction in disease. However, when considering the emergence of resistance, the more important consideration becomes the quality and efficacy of treatment given. If poor treatment is given to an increasing proportion of cases, resistance levels may be exacerbated.

There are numerous examples of resistance emerging in populations in which there may be high rates of treatment, but the quality of treatment is poor in either localized or wide scale regions. In particular, prolonged periods of poor control practices have resulted in rapid development of high rates of resistance. For example, in the Russian Federation, significant resistance has emerged despite high levels of case detection. Overall, an estimated 65% of new smear positive cases are detected, with over 90% treated under a DOTS program. The treatment success rate is 56.7%. Although there remains a reasonably low incidence rate of 23.4 cases per 100,000, there has developed a high prevalence of 32.4% single drug resistance and 9% MDR. A similar situation is found in Latvia, which has achieved 73.3% case detection, with over 90% detected under a DOTS program. With a treatment success rate of 70.2%, the estimated smear positive incidence rate has been reduced to a medium level of 37.1 cases per 100,000. However, there is a high prevalence of 29.9% single drug resistance and 9% MDR. (WHO/IUATLD 1997; Iseman 1999; WHO/IUATLD 2000)

Peru also shows a high prevalence of resistance although control practices have improved considerably over the past decade. Recent program improvements have reached a case detection rate of 81.9%, with over 90% treated under a DOTS program, and an average treatment success rate of 93%. However, a high prevalence of

resistance remains, with 18% single drug resistance and 3% MDR. This high resistance prevalence may reflect earlier poor treatment practices that have allowed the emergence of a continuing resistance epidemic. (Netto, Dye et al. 1999; Espinal, Kim et al. 2000; WHO/IUATLD 2000; Raviglione, Gupta et al. 2001)

V. D. Treatment waiting times

The waiting time to detection and treatment of infectious cases is another important aspect of control. Reducing this waiting time is considered a key element in reducing duration of infectiousness and ongoing cycles of transmission. In these analyses, shortening the average case waiting time resulted in decreased duration of infectiousness, transmission and disease levels. The waiting time to begin treatment was most important in scenarios with a strong first line control program. On top of strong control, including high case detection and first line cure, shortening waiting times results in additional reductions in disease.

In regions with poor control, which have low rates of case detection, reducing waiting times results in slight reductions in disease. However, in this scenario far more disease would be prevented by first increasing case detection. Under programs with low average cure rates, reducing the waiting times may provide even less benefit. Reducing the time to treatment, when treatment itself provides only a marginal reduction in duration of infectiousness, has a limited impact on reducing disease. In this scenario, the priority would lie in first improving control through higher cure rates.

V. E. Reducing waiting time to treatment

The waiting time to treatment can be reduced at several levels. First, new cases must receive medical attention. Second, these patients are seen by a medical practitioner and considered to have symptoms that allow suspicion of a case. Third,

these cases are properly diagnosed as an active case of TB. Fourth, confirmed cases wait to begin a course of treatment.

V. E. 1. delays in patient reporting

The time from onset of symptoms until a patient reports to a health authority is referred to as the patient delay. Studies have considered any time over one month as a delay in patient reporting. Patients may delay seeking for a number of reasons. First, they may have few symptoms and not recognize the need for medical attention. This may occur for biological reasons—a proportion of cases will have lower severity of disease and develop fewer symptoms. Second, there may be social, cultural, or economic barriers to routine medical access. (Asch, Leake et al. 1998; Needham, Foster et al. 2001) In developing country settings, studies have found that delays in patient reporting were correlated to poor health information, and lack of information regarding the symptoms and treatment of tuberculosis. These patient delays were found to be longer in rural areas, in patients with lower education levels, and for patients who did not have prior information regarding tuberculosis. (Salaniponi, Harries et al. 2000; Wandwalo and Morkve 2000; Lienhardt, Rowley et al. 2001; Needham, Foster et al. 2001) Improving general awareness on the symptoms of tuberculosis, available treatment, and how to access health care services are important measures to both increase case detection rates and reduce the delay in patient reporting.

Even under strong control programs in developed countries, there may be significant patient delays in seeking care. Studies examining the reasons patients delay in reporting include concern over cost of treatment, difficulties in access to health services, misunderstanding of tuberculosis, and fearing immigration authorities. Patient delay was correlated to unemployment, uncertainty about where to seek care, and a belief that they could treat themselves. Conversely, the severity of the illness and

symptoms were not seen to significantly affect patient delay. (Asch, Leake et al. 1998) These findings suggest a lack of equity in treatment, such that groups with lower education and knowledge about seeking health care and tuberculosis disease itself are less likely to receive treatment.

Active screening programs, including contact tracing and screening of high-risk groups, may detect these cases and bring them in for treatment faster. In addition, population-wide (or within high-risk groups) measures to increase education about TB symptoms and where to seek treatment may increase detection and reduce reporting times. Education can also seek to reduce possible stigma associated with seeking treatment. Similarly, improving routine medical access, particularly in high-risk groups which often experience social or economic barriers, may decrease reporting times.

V. E. 2. institutional delay in recognizing symptoms / suspecting case

Improvements in education of TB symptoms and diagnosis amongst medical practitioners are of key importance in decreasing the time to suspicion of active TB. This time is often the most critical, as after suspicion many health authorities will seek to isolate patients, or at least reduce potential contacts, until diagnostic tests are returned. Increased medical education is particularly important in areas of low incidence, where practitioners may be unfamiliar with recognizing symptoms and the appropriate guidelines for diagnosis and treatment.

Patients may experience delays once in hospital, waiting either for diagnosis or to start treatment. These delays may include intervals for referral, diagnosis, and initiation of treatment. Both doctor and clinic delays have been shown to contribute to overall institutional delays. (Salaniponi, Harries et al. 2000; Yilmaz, Boga et al. 2001) Clinical delays in treatment are often associated with atypical presentation, lack of

symptoms, and waiting for culture results. Older patients and those without respiratory symptoms are more often initially misdiagnosed. (Mathur, Sacks et al. 1994)

Delays in treatment are associated with admittance to a hospital with a low incidence of tuberculosis, and atypical clinical presentation. In hospitals with low TB admission rates, cases were more likely to be initially misdiagnosed. Further, the rate of TB admissions was inversely proportional to the successful treatment outcome. At lower rates of TB admissions, cases were more likely to fail treatment, die, and transmit infection. Treatment delays amongst patients in a hospital may lead to nosociomal transmission. (Greenaway, Menzies et al. 2002)

V. E. 3. diagnostic delay

Faster and more effective diagnostic tests would allow cases to begin treatment earlier. Diagnostic tests with improved sensitivity may also allow detection of cases at earlier stages, when patients may have fewer disease symptoms. (Heymann, Brewer et al. 1997)

V. E. 4. diagnosis to treatment

Delays from the time of confirmed diagnosis to the start of treatment are usually the shortest, and reflect administration and medical delays. The delay from confirmed diagnosis to treatment tends to range from one to two days in industrialized countries. Significant delays may be caused by administration, referral to specialized centers, or lack of appropriate regimens. In addition, if patients are not hospitalized after diagnosis, further delays may occur if patient does not return as expected.

V. E. 5. An alternative: presumptive treatment

Treatment waiting times can also be reduced by lowering the threshold needed for positive diagnosis. One alternative is starting presumptive treatment without

bacteriological confirmation of mycobacterial infection. Confirmation of disease may come either from subsequent sputum culture test, or simply from patient response to chemotherapy. Such presumptive treatment is used in a variety of situations, including high suspicion of disease, and limited resources for diagnostic testing. (Kennedy and Fallon 1979; Brewer, Heymann et al. 1998)

Reducing times from suspicion of TB to treatment can also be reduced by beginning therapy before confirmation by culture test. In the USA, national guidelines recommend beginning treatment on all patients with positive smear tests. However, studies of practice in hospitals that rarely see active TB cases show that most patients did not begin treatment until after mycobacterial diagnostic confirmation by culture test. This delay, past the requirements of the national guidelines, may reflect on the physician's lack of familiarity with procedures for treating TB cases. Indeed, the time to treatment is faster in hospitals with a higher TB caseload. It remains unclear the effect of treating uninfected patients, and it is believed that this is outweighed by the benefits of reducing time treatment in infectious patients, and reducing the extent of nosociomal transmission. This is less applicable in regions with high environmental and non-tuberculosis mycobacteria, which cannot be distinguished by smear test. In these regions, confirmation by culture test is still needed to signal active TB.

V. F. Drug resistance

V. F. 1. Emergence and control

In the simulations presented here, drug resistance is seen to emerge under first line control programs. As the prevalence of resistance increases, first line control alone is not sufficient to control resistance. Implementation of second line control, on top of strong first line programs, can reduce resistance that may have emerged under earlier

control, and prevent further emergence of resistance. In order to be successful in reducing resistance, the second line control program should include drug testing of a high proportion of cases and offer effective resistance cure.

This is also observed in program conditions. Traditional DOTS programs recommend treating all presenting cases with first line regimens. Poor program practices may allow for the emergence of high levels of resistance. While the first line regimens offered under DOTS may achieve high cure rates for drug sensitive cases, even under the best program conditions, to both INH and RIF (MDR) results in high failure rates. In addition, mistreatment of resistant cases may lead to amplification of a patient's resistance to higher levels. (Mitchison and Nunn 1986; Shimao 1998; Espinal, Kim et al. 2000; Raviglione, Gupta et al. 2001)

The cost and difficulties in implementing wide-scale drug testing and resistance regimens limits the feasibility of these control measures in many parts of the world. However, the good news is that even partial implementation of these second line control measures will still have positive effects. In order to reduce and prevent further resistance, resources available after ensuring high detection and first line cure rates should be invested in second line control.

V. F. 2. DOTS-plus and resistance treatment guidelines

In order to address the growing prevalence of resistance, and in particular MDR, the WHO has considered extending the DOTS program in some regions. The DOTS-plus program incorporates a standardized system for second line control, including drug susceptibility testing, and specialized regimens for treatment of general resistance and MDR. The WHO control guidelines recommend implementation of these additional control measures only in areas that have already achieved targets for DOTS control, maintaining high levels of detection and treatment success. These programs are

particularly important in regions with high prevalence of MDR. Under traditional DOTS, all cases are treated with first line regimens, and the low cure rates of MDR on these regimens may only exacerbate the resistance problem. (Farmer and Kim 1998; WHO 1999; Bastian, Rigouts et al. 2000) (Bahrmand, Velayati et al. 2000) (Loddenkemper, Sagebiel et al. 2002)

Regions that lack the resources for wide-scale drug testing and expensive MDR treatments, but do have a growing resistance problem, have other alternatives. Some programs offer a general resistance treatment by default: cases failing one or more subsequent courses of treatment receive a standardized re-treatment regimen that shows higher cure rates for some resistant cases. (WHO 1997; Bahrmand, Velayati et al. 2000; Loddenkemper, Sagebiel et al. 2002)

VI. Summary of Findings

In this chapter, the optimal control strategies for regional TB epidemics were explored, incorporating historical trends in TB treatment and following the course of epidemics through incremental improvements in control programs, including both first and second line chemotherapy. The effects of introducing elements of first and second line control programs on the dynamics of the epidemic, as well as the benefits in terms of reducing disease and preventing resistance, were compared.

While treatment will generally reduce disease levels, this depends upon the efficacy of treatment and the probability of acquiring resistance. Even treatment with low cure rates will reduce disease, by reducing the case DOI below that of untreated cases. However, if there is a high probability of acquiring resistance, then increasing treatment under low cure rates will select for higher levels of resistance and possibly disease.

Treatment also brings a cost by selecting for drug resistance. This selection is greater under higher treatment and lower cure rates. Treating a high proportion of cases with low cure rates will allow for the emergence and spread of resistance. If low cure rates are maintained while treatment coverage is expanded, it is possible to develop a serious resistance problem, in which additional treatment leads to greater resistance and possibly disease.

Introducing a second line treatment program, including the drug testing and specialized treatment regimens, reduces prevalence of both disease and resistance. The success of a second line treatment program depends upon the proportion of cases receiving drug susceptibility treatment and the efficacy of resistance treatment, as well as the strength of the first line treatment program, including the case detection and (drug-sensitive case) cure rates.

9. Chapter 9

Control priorities

9. CHAPTER 9
CONTROL PRIORITIES
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IX. CONCLUSIONS

I. Goals

I. A. To consider control priorities for TB epidemics in high and low disease burden settings.

II. Abstract

The control measures outlined in the previous chapter are further explored by examining the benefits of implementing these measures within different settings. The relative benefits of different control measures are considered and priorities for the development of control programs recommended. Current global targets for increasing

case detection and cure rates are appropriate for the high burden countries, where over 80% of cases are found. In these conditions, the control of epidemics is hindered by low case detection and treatment rates. In regions where a high prevalence of resistance has already developed, special care must be taken to improve program conditions before considering expansion. In low incidence regions, control priorities can expand to include additional measures, specifically, active case detection and treatment of latently infected individuals.

III.Introduction

In this chapter, the relative priorities in control of local TB epidemics are considered. Three representative settings were selected that differ by disease burden, level of control, and resistance prevalence. The relative benefits of implementing elements of first and second line control on reducing incidence of disease and resistance were tested.

These settings are:

1. **A high burden country with low resistance prevalence and strong 1st line control but low detection rates.**
2. **A high burden country with poor treatment practices, high rates of acquired resistance, and weak 1st line control.**
3. **A low incidence setting with medium levels of resistance and strong 1st line but no 2nd line control.**

The disease burden and level of control simulated in each of these three scenarios is summarized in Table 9-1. (Table 9-1)

Scenario	1	2	3
	HBC Low resistance	HBC High resistance	Low incidence, elimination phase
Disease burden			
Incidence rate	62	85	21

Incidence rate DR	4.5	26	1.5
Prevalence DR	9%	33%	6.7%
Control program			
CDR	40%	40%	80%
Cure rate, DS	60%	40%	80%
DST	0%	0%	0%

Table 9-1. Summary of disease burden and control parameters used in the three scenarios described in this chapter.

IV. Scenario 1: High Burden

IV. A. Description scenario 1

The first scenario reflects a high burden country with a low level of resistance. The disease levels are high, with an annual incidence rate of 62.5 cases per 100,000. Resistance is at a low level, with under 10% resistance prevalence amongst cases. A first line control program, with a case detection rate of 40% and treatment cure rate of 60%, has been maintained for twenty years.

IV. B.Trends

In these simulations, an epidemic was first begun by introducing a drug sensitive case into a susceptible population of size 10^8. After 100 years in the absence of treatment, the epidemic reached equilibrium levels of disease with an annual incidence rate of 163 cases per 100,000. Control was then introduced with a case detection rate of 40% and an average cure rate of 60%. Under 20 years of this control, disease levels were reduced to an annual incidence of 62.5 cases per 100,000. This control program also led to an increased resistance prevalence of 8.7% (resistance incidence of 4.5 cases per 100,000). (Figure 9-1)

If this control program is continued for an additional 10 years, the disease prevalence will continue to decrease to an incidence rate of 61 cases per 100,000. The

resistance prevalence will also continue to increase to reach resistance incidence rate of 6.5 cases per 100,000 (12% resistance prevalence). (Figure 9-1) If the control program is improved over the next 10 years of control, through increased case detection and/or treatment cure rates, the prevalence of disease will be further reduced.

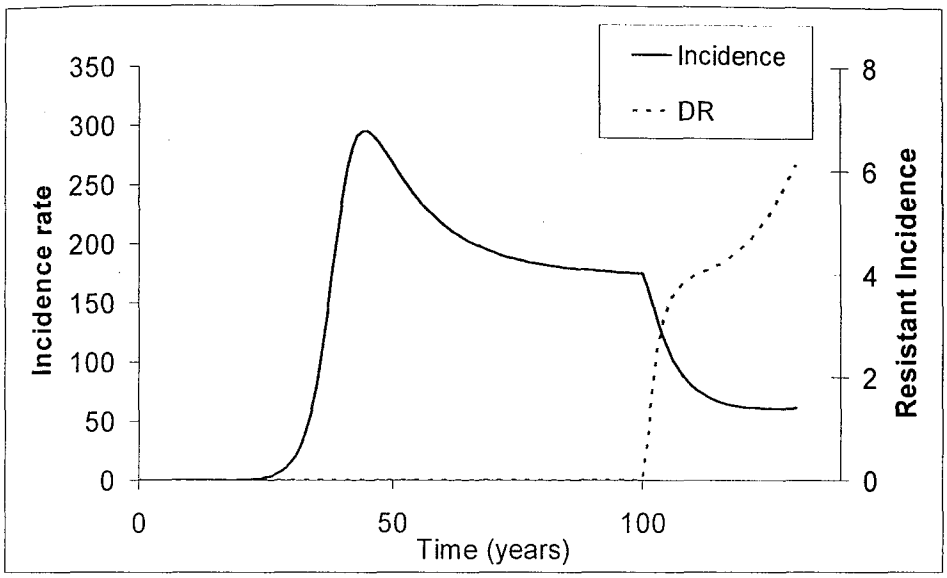


Figure 9-1. ***Trends in incidence for simulations in scenario 1.***
Simulations were run as described in the text for Scenario 1. For the first 100 years of the simulation, the epidemic was allowed to equilibrate in the absence of treatment, reaching final annual incidence rate of 163 cases per 100,000. For years 101- 120, a control program including case detection rate of 40% and cure rate of 60% was implemented. For the final years 121-130, this same control program was maintained. This control reduced the incidence rate to 61 cases per 100,000, but allows for an increase in resistance incidence to 6.5 cases per 100,000. Shown is the annual incidence rate of total and resistant cases (as cases per 100,000) at the end of the simulation.

IV. C. Relative Priorities

The control parameter that had the greatest influence on the prevalence of disease was the case detection rate. Increasing the case detection rate from 40% to 50% resulted in a reduction in annual incidence from 63.6 to 53.8 cases per 100,000. A further increase to 60% case detection resulted in a further decrease to 45.9 cases per 100,000. (Figure 9-2, Table 9-1) Increasing the case detection rate resulted in reduced incidence of drug sensitive cases. Increasing the case detection rate to 60% reduced the

incidence of sensitive cases from 55.2 to 38.1 cases per 100,000. The incidence of resistant was slightly reduced from 8.4 to 7.9 cases per 100,000. (Figure 9-2, Table 9-2)

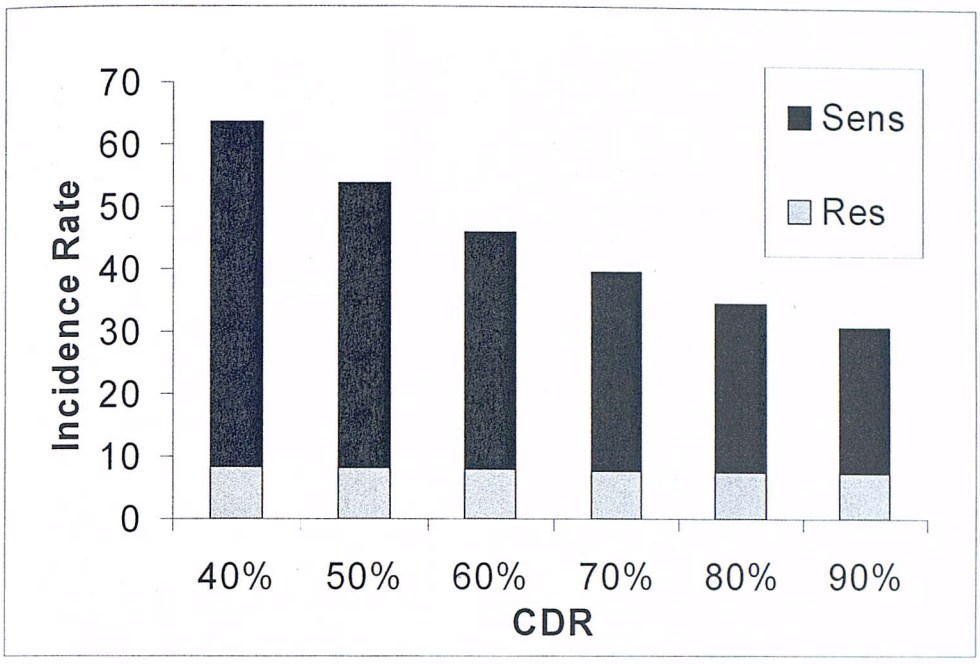


Figure 9-2. ***Increases to the case detection rate reduce the incidence of drug sensitive cases.*** Increasing the case detection rate from 40% to 90% resulted in a reduction in incidence of sensitive cases. Simulations were run as described in the text for Scenario 1. During the final ten years of the simulations, the case detection rate was set at values from 40% to 90%, while the treatment cure rate was held constant at 60%. No drug testing or resistance treatment was included. Shown is the annual incidence rate of drug sensitive and resistant cases (as cases per 100,000) at the end of the simulation.

The control parameter with the second greatest influence on disease levels was the cure rate. Increasing the average treatment cure rate from 60% to 80% reduced the annual incidence from 63.6 to 55.1 cases per 100,000. Increasing the cure rate to 90% reduced the annual incidence to 51.9 cases per 100,000. As shown in Figure 9-3, increasing the case detection and cure rates simultaneously results in further decreases in incidence. (Figure 9-3)

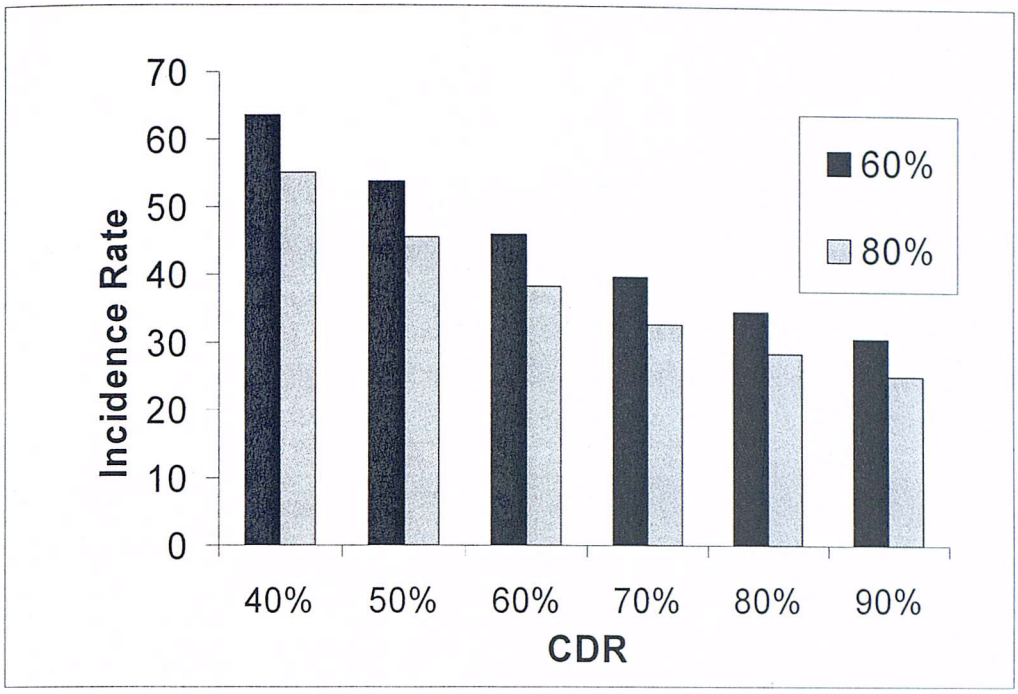


Figure 9-3. ***Increases to the case detection and cure rates reduce incidence.*** Higher treatment and cure rates result in lower rates of incidence. Simulations were run as described in the text for Scenario 1. During the final ten years of the simulations, the case detection rate was set at values from 40% to 90%, while the treatment cure rate was held constant at either 60% or 80%. No drug testing or resistance treatment was included. Shown is the annual incidence rate (as cases per 100,000) at the end of the simulation.

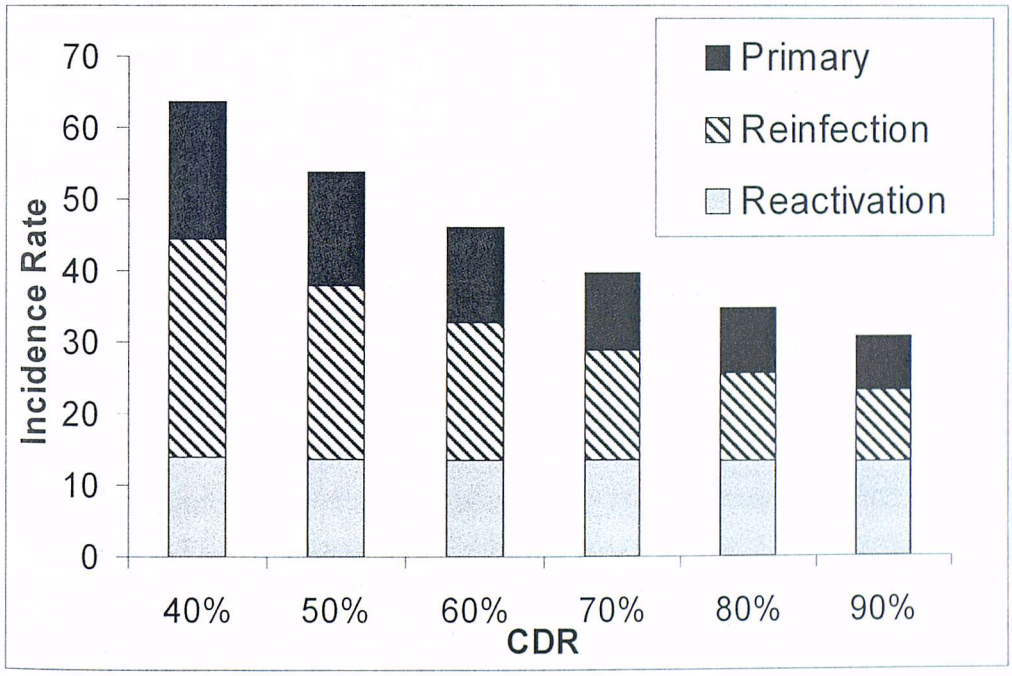


Figure 9-4. ***Increases to the case detection rate reduce the incidence of cases caused by primary disease and reinfection.*** Higher levels of case detection and treatment reduce the incidence of primary and reinfection cases, but leave nearly constant the incidence of reactivation cases. Simulations were run as described in the text for Scenario 1. During the final ten years of the simulations, the case detection rate was set at values from 40% to 90%, while the treatment cure rate was held constant at 60%. No drug testing or resistance

treatment was included. Shown is the annual incidence rate (as cases per 100,000) at the end of the simulation, by type of case. Reactivation cases are due to reactivation of an earlier infection, primary cases are due to direct progression after infection, and reinfection is due to direct progression after reinfection.

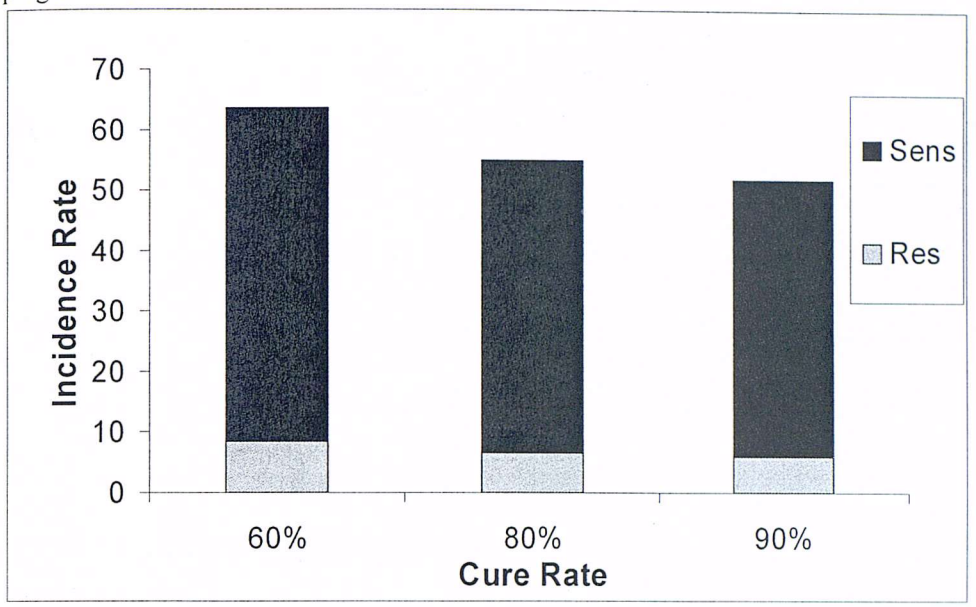


Figure 9-5. ***Increasing the treatment cure rate for sensitive cases reduces the incidence of drug sensitive and resistant cases.*** The improved treatment cure rates reduce the incidence of both drug sensitive and resistant as cases. Simulations were run as described in the text for Scenario 1. During the final ten years of the simulations, the first line cure rate was set at values from 60% to 90%, while the case detection rate was held constant at 40%. No drug testing or resistance treatment was included. Shown is the annual incidence rate of drug sensitive and resistant cases (as cases per 100,000) at the end of the simulation.

The first-line control parameter that had the greatest influence on the resistance prevalence was the cure rate. Increasing the cure rate from 60% to 80% (while keeping the case detection rate at 40%) reduced the annual incidence of resistant cases from 8.4 to 6.5 cases per 100,000. Increasing the cure rate to 90% further reduced resistance to 5.9 cases per 100,000. (Figure 9-5, Table 9-2) Other first-line control measures had comparatively low impact on disease levels and resistance prevalence.

Introducing drug testing and resistance treatment resulted in only slight reductions in resistance. Introducing drug testing and resistance treatment of 50% of incident cases resulted in a reduction of annual incidence of resistance cases from 8.4 to 8.2 cases per 100,000. Testing of a higher proportion of incident cases resulted in further decreases in incidence of resistance cases. (Table 9-2) However, in order to

achieve significant reduction of overall disease, as well as the remaining resistance, the case detection and cure rates must also be increased. For example, increasing the case detection rate to 60%, the DS cure rate to 80%, and the DST rate to 100%, the resistance incidence rate is reduced to 4.8 cases per 100,000. This increase in DST does not significantly affect the incidence of sensitive cases, which stays near 32 cases per 100,000. (Figure 9-5, Table 9-2) Further reductions to remaining resistance require higher levels of case detection, in order to find and treat new resistant cases, and to reduce the pool of sensitive cases from which new resistance arises.

Introducing drug testing resulted in a reduction in the incidence of resistance amongst primary cases, but left that amongst reactivation cases nearly constant. (Figure 9-6) This shows that the instituted drug testing and resistance treatment reduced the ongoing transmission of resistance, lowering the incidence of resistance amongst primary cases.

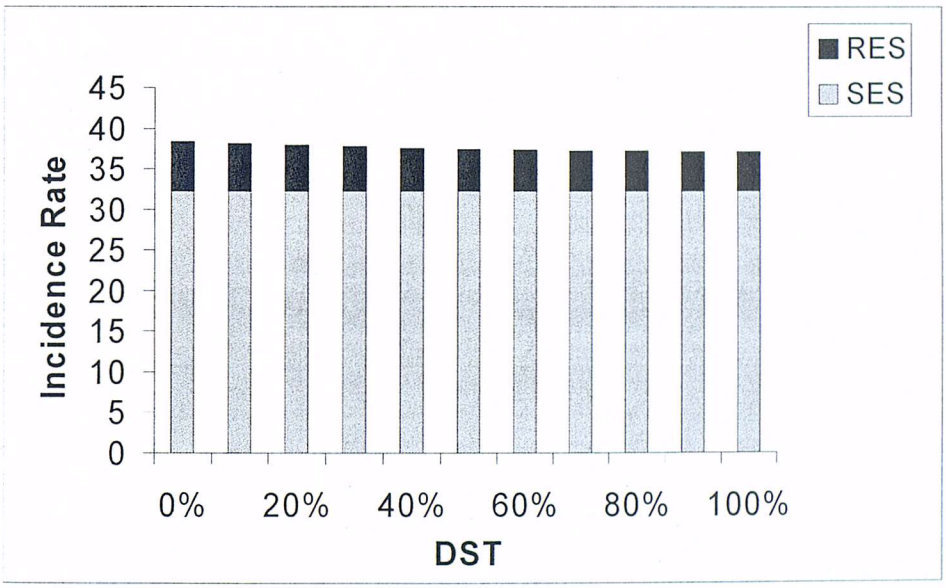


Figure 9-6. ***Increases to the drug susceptibility testing rate (DST) reduces the incidence of resistant cases.*** Simulations were run as described in the text for Scenario 1. During the final ten years of the simulations, the case detection rate was set at 60%, the cure rate for DS cases at 80%. The proportion of incident cases receiving DST and resistance treatment was set at values between 0% and 100%. The cure rate for singly resistant and MDR cases is set at 60% and 40%, respectively. Shown is the annual incidence rate (as cases per 100,000) at the end of the simulation, by resistance status.

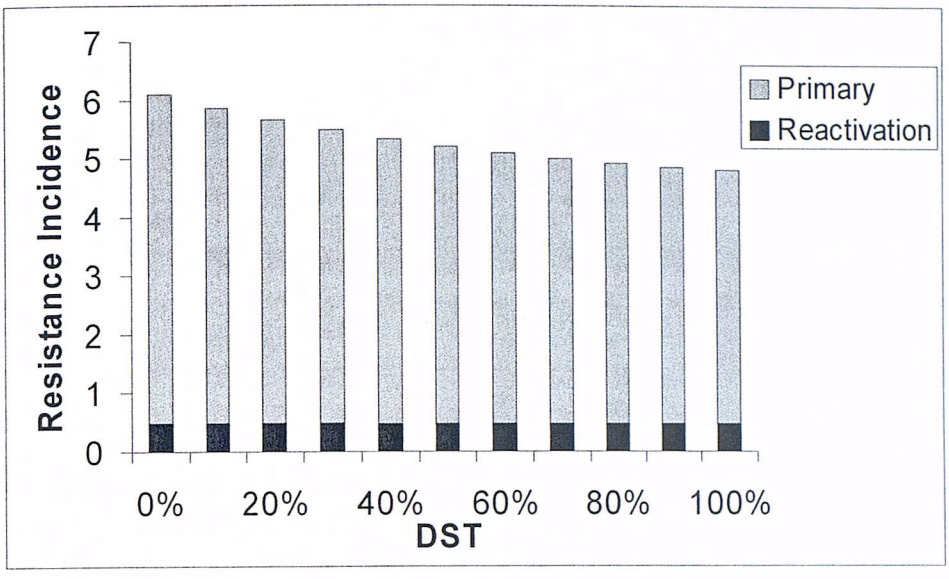


Figure 9-7. ***Increases to the DST rate reduce the incidence of resistant cases caused by primary disease.*** Simulations were run as described in the text for Scenario 1. During the final ten years of the simulations, the case detection rate was set at 60% and cure rate (of drug sensitive cases) was set at 80%. The drug testing rate of new incident cases was set at values between 0% and 100%, as shown. The cure rate for singly resistant and MDR cases is set at 60% and 40%, respectively. Shown is the annual incidence rate (as cases per 100,000) at the end of the simulation, by type of case. Reactivation cases are due to reactivation of an earlier infection, primary cases are due to direct progression after infection.

Shortening the average case waiting time to detection and treatment also provided a small reduction in incidence. The waiting time to treatment was set at 4 months during the first twenty years of control in the simulations shown in Figure 9-7. During the final ten years of control, the waiting time was reduced to values between 1 and 3.5 months. Reducing the treatment waiting time from 4 months to 1 month resulted in a decrease in incidence rate from 65 to 60 cases per 100,000. While the incidence of drug sensitive cases was reduced from 57.2 to 51.8, the drug resistant incidence rate stayed fairly constant at near 8.2 cases per 100,000. (Figure 9-8) While shortening the time to treatment does slightly reduce the incidence of disease, the benefits are small compared to other measures. A higher case detection and cure rates are needed before treatment waiting times has a more significant impact in preventing disease.

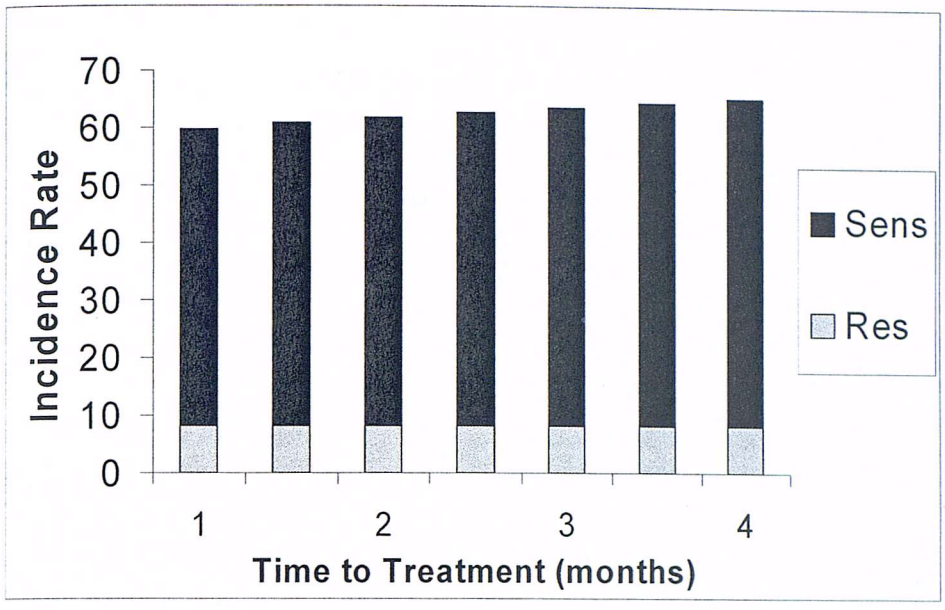


Figure 9-8. ***Reducing the time to treatment reduces incidence.*** Shortening the average waiting time to case treatment from 4 months to 1 month reduced the incidence of sensitive cases from 57.2 to 51.8 cases per 100,000, while the incidence of resistance was only slightly reduced from 8.2 to 8.1 cases per 100,000.

Simulations were run as described in the text for Scenario 1. During the first twenty years of control, the waiting time to treatment was set at 4 months. The treatment waiting time was either kept at 4 months, or reduced to values from 1 to 3.5 months for the final ten years of the simulations. Throughout the simulations, the case detection rate was set at 40% and the treatment cure rate at 60%. No drug testing or resistance treatment was included. Shown is the annual incidence rate (as cases per 100,000) of drug sensitive and resistant cases at the end of the simulation.

V. Scenario 2: High burden region, high resistance

V. A. Description

The second scenario presented here is of a high burden country with a high prevalence of resistance. In these simulations, there is an annual incidence rate of near 100 cases per 100,000, with a resistance prevalence of 33%. There is a first line control program in place, with a case detection rate of 40% and cure rate of 40%. There is no second line control in place. Treatment practices and low cure rates have allowed for the emergence of high rates of resistance, which are not effectively treated under the current program. In addition, the ability to control the still rising epidemic is limited by a low case detection rate. This scenario reflects those HBCs which have medium to

low first line control practices, with variable treatment efficacy and practices, and have developed a high prevalence of resistance.

V. B. Trends

In these simulations, an epidemic was first run to equilibrium levels of disease, reaching an annual incidence rate of 280 cases per 100,000 after 100 years. Control was then introduced, with a case detection rate of 40% and cure rate of 40%. This control was maintained for twenty years, reducing incidence to 85 cases per 100,000. This control also resulted in an increase in incidence of resistance to 26 cases per 100,000 (a resistance prevalence of 33%). In the final ten years of the simulations, this control program was improved in some way, and the results of these simulations are shown in the tables below. (Figure 9-9)

First we examine the impact of maintaining the same control program for the final ten years of the simulation, with a case detection rate of 40% and a cure rate of 40%, and no drug testing or resistance treatment. This results in a continued decrease in drug sensitive cases to an annual incidence rate of 53 cases per 100,000. The resistance rate, at the same time, will continue to increase to an incidence rate of approximately of 37 cases per 100,000 (a resistance prevalence of 41%). Due to this continued growth in resistance, the total incidence rate has increased slightly to 90 cases per 100,000. (Figure 9-9) Improving the control, over the final ten years of the simulation, results in further decreases in disease as well as resistance.

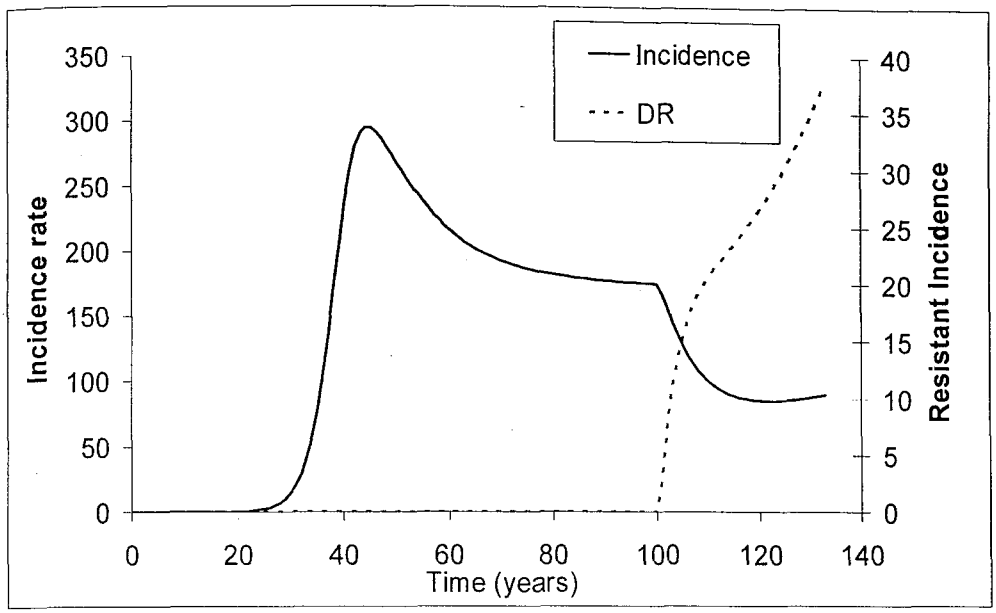


Figure 9-9. ***Trends in incidence for simulations in scenario 2.*** Simulations were run as described in the text for Scenario 2. For the first 100 years of the simulation, the epidemic was allowed to equilibrate in the absence of treatment, reaching final annual incidence rate of 170 cases per 100,000. For years 100- 130, a control program including case detection rate of 40% and cure rate of 40% was implemented. This reduces the total incidence rate to 85 cases per 100,000, while allowing an increase of resistance incidence to 26 cases per 100,000. For the final years 131-140, this same control program was maintained. This continued control reduces the drug sensitive incidence rate to 53 cases per 100,000, while allowing the resistance incidence to increase to 37 cases per 100,000, combining to a slightly increased total incidence of 90 cases per 100,000. Shown is the annual incidence rate of total and resistant cases (as cases per 100,000) at the end of the simulation.

V. C. Relative Priorities

The control parameter that had the greatest influence on the prevalence of disease was the case detection rate. Increasing the case detection rate from 40% to 50% resulted in a reduction in annual incidence from 90.2 to 81.9 cases per 100,000. A further increase to 60% case detection reduced incidence to 75.4 cases per 100,000. (**Figure 9-10**) Comparing the numbers of sensitive and resistant amongst the new incident cases in **Figure 9-10**, shows that increasing the case detection rate reduced the incidence of drug sensitive cases, while allowing resistance to increase. Increasing the case detection rate to 60% reduced the incidence of sensitive cases from 52.4 to 36.5 cases per 100,000. At the same time, the incidence of resistant cases increased slightly from 37.8 to 38.8 cases per 100,000. (Figure 9-10, Table 9-3) Taking another look at the components of new incident cases in **Figure 9-11**, shows that increasing the case

detection rate reduces the incidence of primary and reinfection cases, but leaves nearly constant the incidence of reactivation cases. (**Figure 9-11**, Table 9-3)

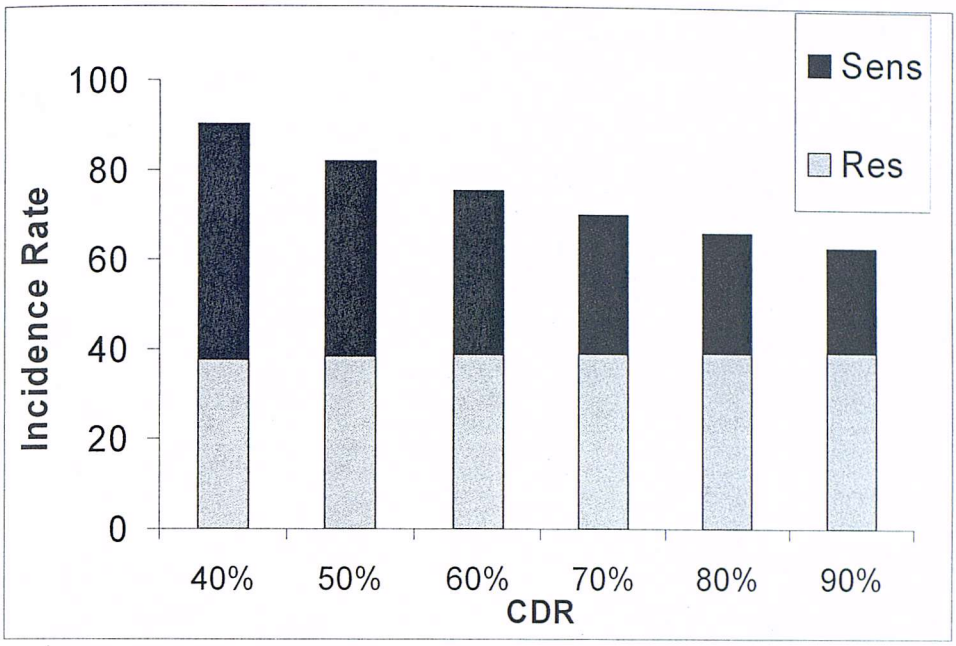


Figure 9-10. ***Increases to the case detection rate reduce the incidence of drug sensitive cases.*** Higher rates of case detection reduced the incidence of drug sensitive cases, while slightly increasing the resistance incidence. Simulations were run as described in the text for Scenario 2. During the final ten years of the simulations, the case detection rate was set at values from 40% to 90%, while the treatment cure rate was held constant at 40%. No drug testing or resistance treatment was included. Shown is the annual incidence rate of drug sensitive and resistant cases (as cases per 100,000) at the end of the simulation.

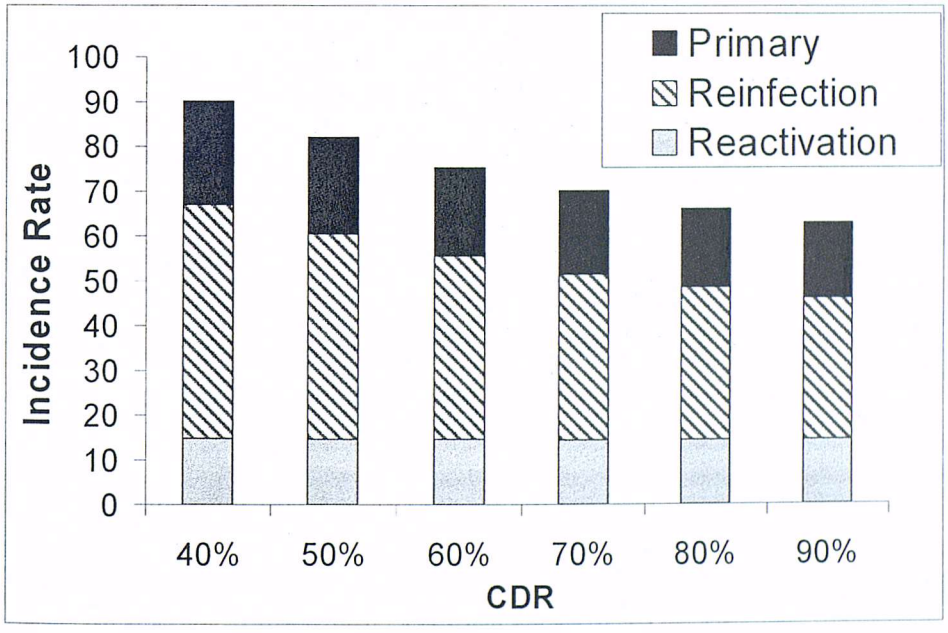


Figure 9-11. ***Increases to the case detection rate reduce the incidence of cases caused by primary disease and reinfection.*** Higher levels of case detection

and treatment reduce the incidence of primary and reinfection cases, but leave nearly constant the incidence of reactivation cases. Simulations were run as described in the text for Scenario 2. During the final ten years of the simulations, the case detection rate was set at values from 40% to 90%, while the treatment cure rate was held constant at 40%. No drug testing or resistance treatment was included. Shown is the annual incidence rate (as cases per 100,000) at the end of the simulation, by type of case. Reactivation cases are due to reactivation of an earlier infection, primary cases are due to direct progression after infection, and reinfection is due to direct progression after reinfection.

The control parameter with the second greatest influence on disease levels was the cure rate. When the average treatment cure rate was increased from 60% to 70%, the annual incidence was reduced from 97.2 to 89.6 cases per 100,000. Increasing the cure rate to 80% or 90% further decreased the annual incidence to 83.4 or 78.3 cases per 100,000, respectively. (Figure 9-12)

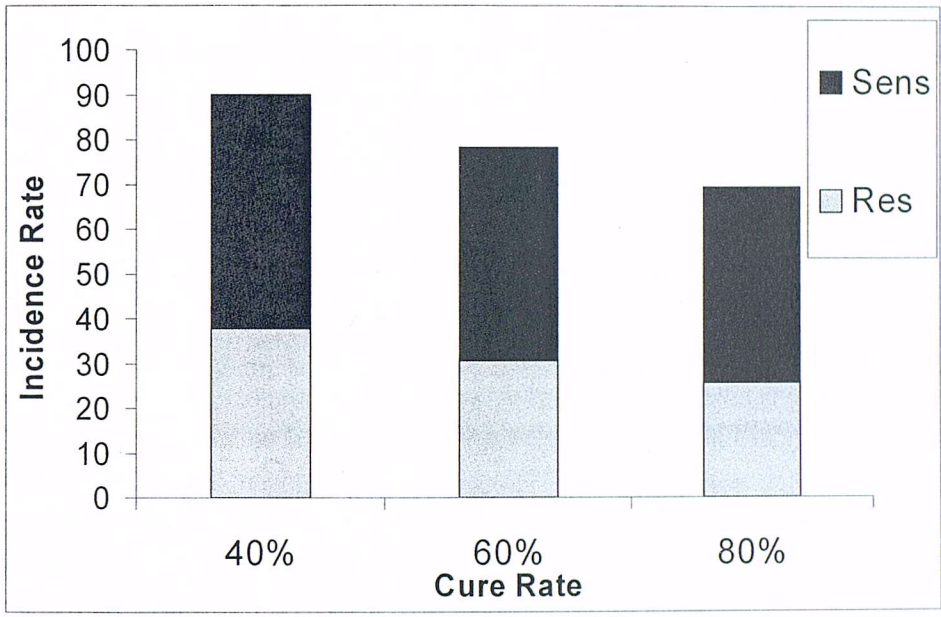


Figure 9-12. ***Increases to the treatment cure rate for sensitive cases reduce the incidence of drug sensitive cases.*** Increasing the cure rates for first line treatment reduces the incidence of both drug sensitive and resistant cases. Simulations were run as described in the text for Scenario 2. During the final ten years of the simulations, the first line cure rate was set at values from 40% to 80%, while the case detection rate was held constant at 40%. No drug testing or resistance treatment was included. Shown is the annual incidence rate of drug sensitive and resistant cases (as cases per 100,000) at the end of the simulation.

The first-line control parameter with the greatest influence on resistance was the cure rate. Increasing the cure rate from 40% to 80% (while keeping the case detection rate at 40%) reduced the annual incidence of resistant cases from 37.8 to 25.5 cases per

100,000. (Figure 9-12, Table 9-3) Other first-line control measures had comparatively low impact on disease levels or resistance prevalence.

V. D. Importance of DST and resistance cure

In this scenario, introducing drug testing and resistance treatment under the current first line control program results in a slight increase in resistance. When drug testing is introduced on top of the current first line program with a CDR of 40% and DS cure rate of 40%, introducing drug testing increases resistance; when 50% of treated cases receive drug testing the incidence of resistance is increased from 37.8 to 40.3 cases per 100,000. (Figure 9-13, Table 9-3)

In this scenario, the case detection and cure rates are too low for widespread drug testing to be beneficial. In addition, there may be a danger in offering second line treatments when there are low cure rates for both sensitive and resistant cases. Introducing second line drugs with regimens that have low cure rates allows for further selection for higher levels of resistance, as well as increasing the number of drugs to which cases are resistant.

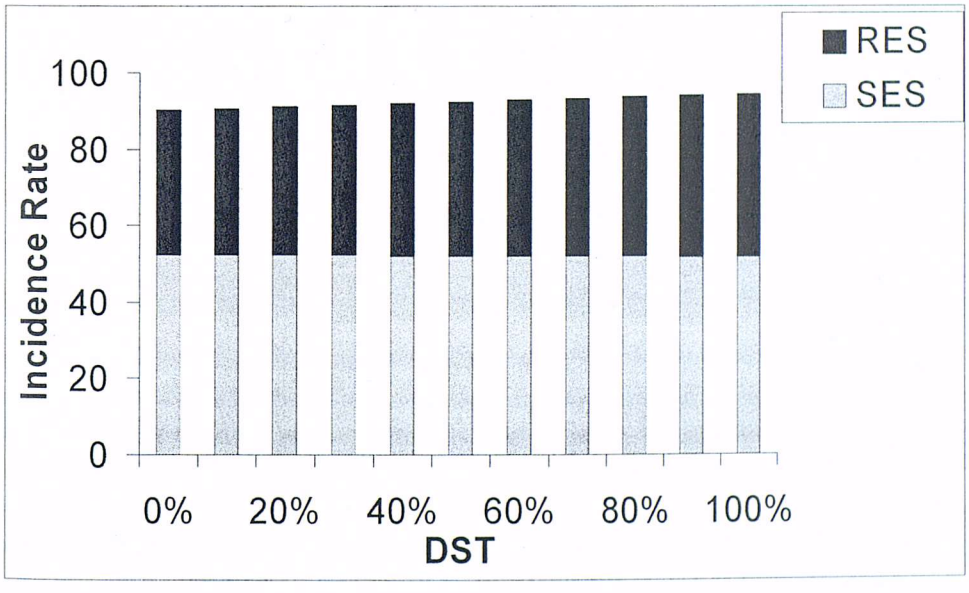


Figure 9-13. ***Increases to the drug susceptibility testing rate (DST) reduces the incidence of resistant cases.*** Simulations were run as described in the text

for Scenario 2. During the final ten years of the simulations, the case detection rate was set at 40%, the cure rate for DS cases at 40%. The proportion of incident cases receiving DST and resistance treatment was set at values between 0% and 100%. The cure rate for singly resistant and MDR cases is set at 40% and 30%, respectively. Shown is the annual incidence rate (as cases per 100,000) at the end of the simulation, by resistance status.

The proportion of resistance found in primary and reactivation cases is next examined. The implementation of drug testing and resistance treatment allows for an increased incidence of resistance amongst primary cases, but does not significantly affect that amongst reactivation cases. When 100% of incident cases receive drug testing, the incidence of resistance amongst primary disease cases is increased from 35.9 to 40.6 cases per 100,000. The incidence of resistance amongst reactivation cases is increased slightly from 1.9 to 2.0 cases per 100,000. (Figure 9-14, Table 9-3) In these simulations, the inclusion of drug testing has increased the ongoing transmission of resistance, as reflected by the increase in resistance incidence amongst primary cases.

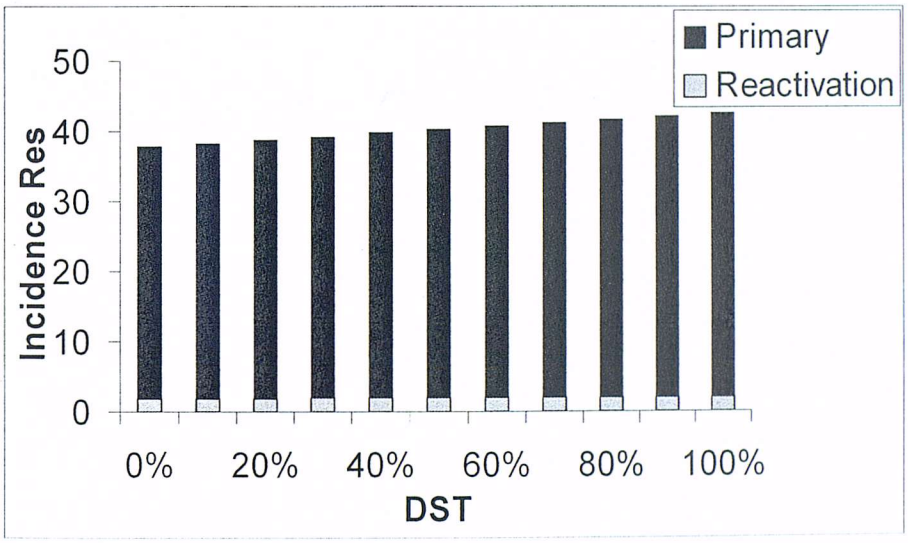


Figure 9-14. ***Increases to the DST rate reduce the incidence of resistant cases caused by primary disease.*** Simulations were run as described in the text for Scenario 2. During the final ten years of the simulations, the case detection rate was set at 40% and cure rate (of drug sensitive cases) was set at 40%. The drug testing rate of new incident cases was set at values between 0% and 100%, as shown. The cure rate for singly resistant and MDR cases is set at 40% and 30%, respectively. Shown is the annual incidence rate (as cases per 100,000) at the end of the simulation, by type of case. Reactivation cases are due to reactivation of an earlier infection, primary cases are due to direct progression after infection.

Shortening the average case waiting time to detection and treatment also provided a small reduction in incidence. The waiting time to treatment was set at 4 months during the first twenty years of control in the simulations shown in **Figure 9-15**. During the final ten years of control, the waiting time was reduced to values between 1 and 3.5 months. Reducing the treatment waiting time from 4 months to 1 month resulted in a decrease in incidence rate from 90 to 85 cases per 100,000. While the incidence of drug sensitive cases was reduced from 52.4 to 47.6, the drug resistant incidence rate stayed fairly constant at near 38 cases per 100,000. (**Figure 9-15**) While shortening the time to treatment does slightly reduce the incidence of disease, the benefits are small compared to other measures. A higher case detection and cure rates are needed before treatment waiting times has a more significant impact in preventing disease.

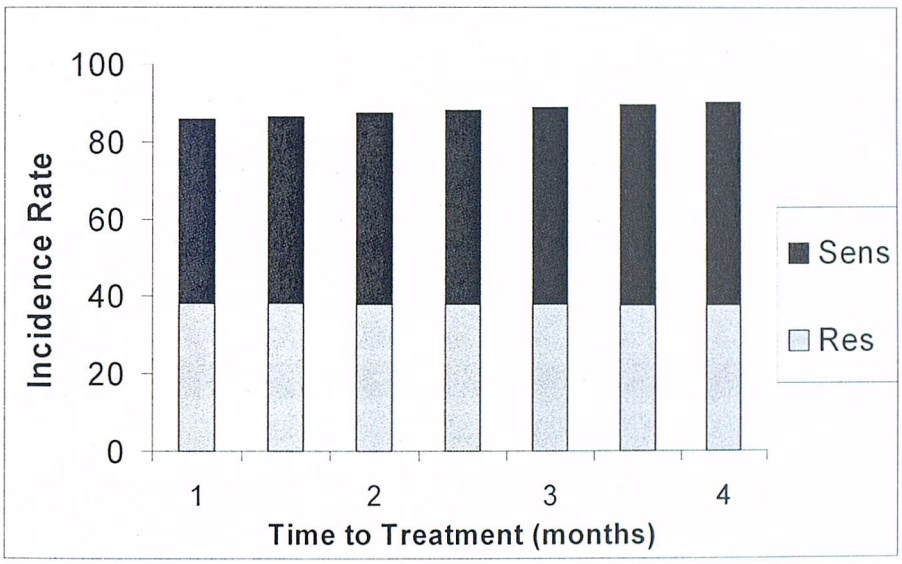


Figure 9-15. ***Reducing the time to treatment reduces incidence.***
Simulations were run as described in the text for Scenario 2. During the first twenty years of control, the waiting time to treatment was set at 4 months. The treatment waiting time was either kept at 4 months, or reduced to values from 1 to 3.5 months for the final ten years of the simulations. Throughout the simulations, the case detection rate was set at 40% and the treatment cure rate at 40%. No drug testing or resistance treatment was included. Shown is the annual incidence rate (as cases per 100,000) of drug sensitive and resistant cases at the end of the simulation.

VI. Scenario 3: Low Incidence Region

VI. A. Description

This scenario reflects a low burden country, with an annual incidence rate below 20 cases per 100,000. The resistance prevalence is at a medium level of 5-15%. There is a strong first line control program, with a case detection rate of 80% and cure rate of 80%, but no second line control in place. The first line control program is strong and has reduced disease to low levels; however, if resistance remains unchecked will continue to increase.

VI. B. Trends

For the first 100 years of the simulations shown in this section, an epidemic was begun by introducing one drug sensitive case into a susceptible population, and allowed to run to equilibrium, reaching an annual incidence rate of 45 cases per 100,000. Control was then introduced, with a case detection rate of 80% and cure rate of 80%, and maintained for thirty years. This control reduced the incidence to 20 cases per 100,000, while allowing for an increase in resistance to an incidence of 2.5 cases per 100,000 (a resistance prevalence of 6.7%). In the final ten years, this control program was improved in some way, and the results of these simulations are shown in the following figures and tables.

First we examine the impact of maintaining the same control program for the final ten years of the simulation, with a case detection and cure rates each at 80%, and no drug testing or resistance cure. This results in a continued decrease in drug sensitive cases to an annual incidence rate of 18 cases per 100,000. The resistance rate continues to increase to an incidence rate of approximately 2.5 cases per 100,000 (a resistance prevalence of 20%). Due to growing resistance, the total incidence rate has slightly increased over the course of these ten years of continued treatment, to reach 21 cases

per 100,000. (**Figure 9-16**) Improving the control, over the final ten years of the simulation, results in further decreases in disease as well as resistance.

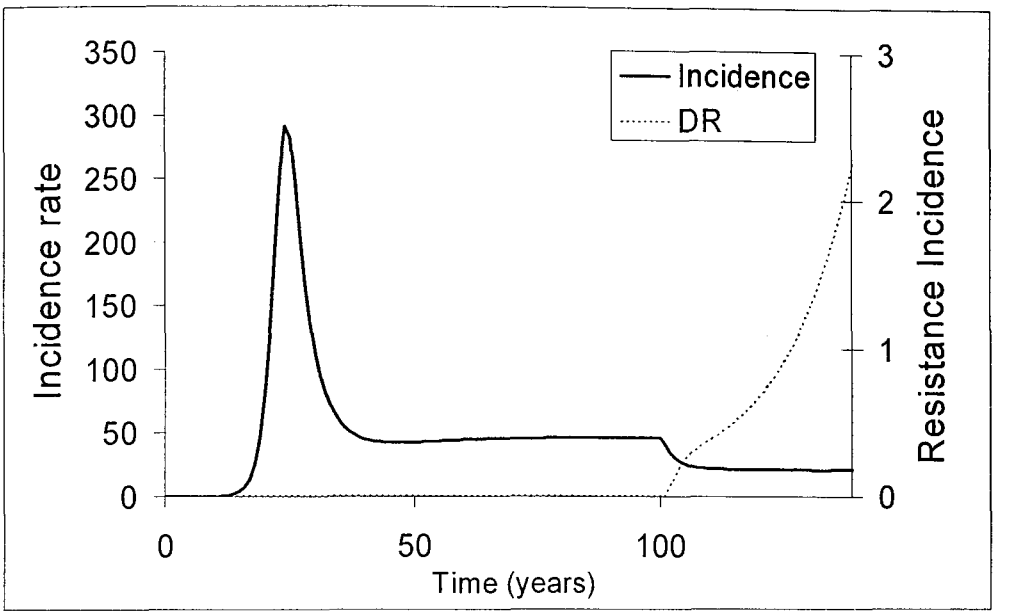


Figure 9-16. ***Trends in incidence for simulations in scenario 3.***
Simulations were run as described in the text for Scenario 3. For the first 100 years of the simulation, the epidemic was allowed to equilibrate in the absence of treatment, reaching final annual incidence rate of 46 cases per 100,000. For years 100- 130, a control program including case detection and cure rates each of 80% was implemented. This reduces the incidence rate to 20.8 cases per 100,000, but allows for an increase in resistance incidence to 1.3 cases per 100,000. For the final years 131-140, this same control program was maintained. This control reduces the incidence rate of drug sensitive cases to 18.6 cases per 100,000, but allows for an increase of resistance to 2.4 cases per 100,000, resulting in a slightly increased combined incidence rate of 21.3 cases per 100,000. Shown is the annual incidence rate of total and resistant cases (as cases per 100,000) at the end of the simulation.

VI. C. Relative Priorities

The control parameter that had the greatest influence on the prevalence of disease was the case detection rate. Increasing the case detection rate from 80% to 90% resulted in a reduction in annual incidence from 21.3 to 18.9 cases per 100,000. The final incidence of sensitive cases was reduced from 18.6 to 16.1, while the resistance incidence stayed nearly constant at 2.7 cases per 100,000. (Figure 9-17)

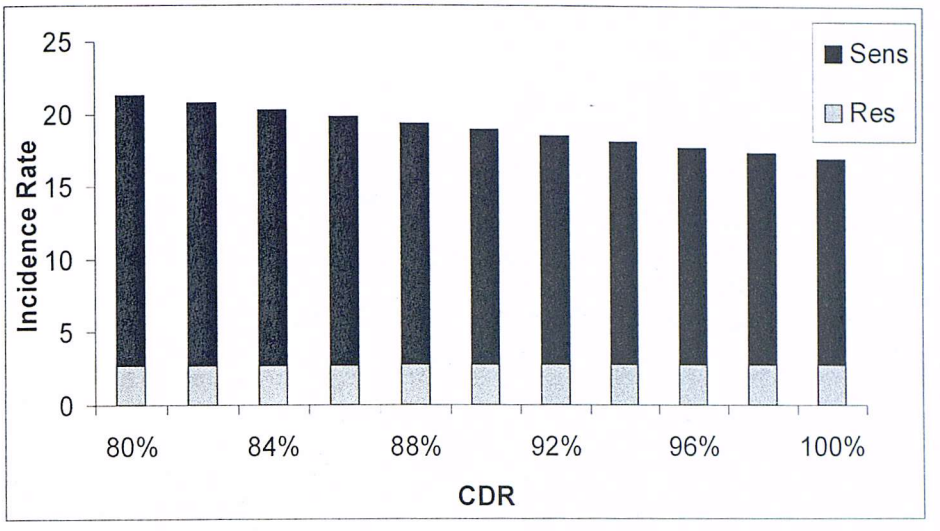


Figure 9-17. ***Increases to the case detection rate reduce the incidence of drug sensitive cases.*** Simulations were run as described in the text for Scenario 3. During the final ten years of the simulations, the case detection rate was set at values from 80% to 100%, while the treatment cure rate was held constant at 80%. No drug testing or resistance treatment was included. Shown is the annual incidence rate of drug sensitive and resistant cases (as cases per 100,000) at the end of the simulation.

Increases to the cure rate, above the already high average of 80% treatment success, results in further decreases in disease levels. As shown in Figure 18, increasing the cure rate to 90% (while keeping the CDR at 80%) reduces the disease incidence from 21.3 to 19.5 cases per 100,000. (Figure 9-18) It is further apparent from Figure 18 that increases to the already high case detection and cure rates do not result in the quick eradication of remaining levels of disease. Even at the highest levels of detection and cure (of drug sensitive cases), there remain a fairly constant level of annual incidence.

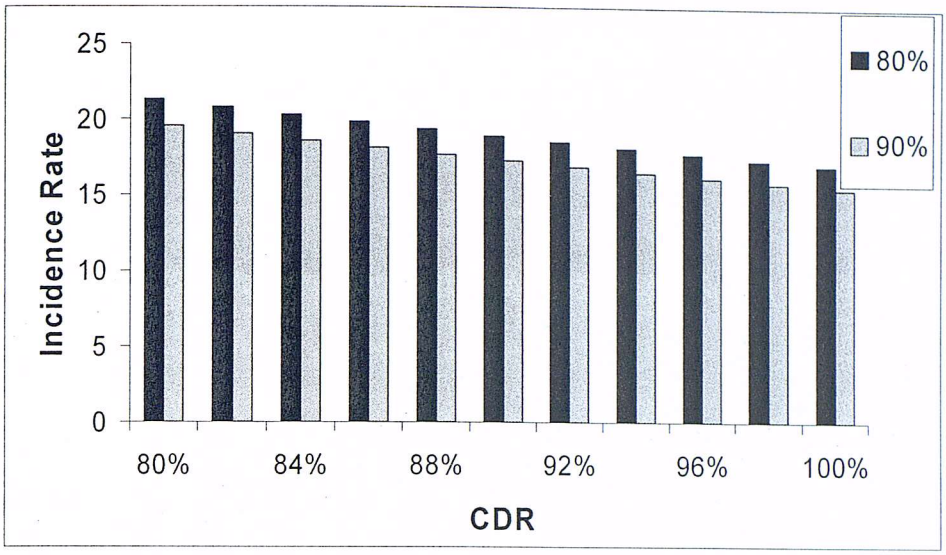


Figure 9-18. ***Increases to the case detection and cure rates reduce incidence.*** Simulations were run as described in the text for Scenario 3. During the final ten years of the simulations, the case detection rate was set at values from 80% to 100%, while the treatment cure rate was held constant at either 80% or 90%. No drug testing or resistance treatment was included. Shown is the annual incidence rate (as cases per 100,000) at the end of the simulation.

(1) Relative importance of primary and reactivation disease

In these simulations, increasing the case detection rate even to very high levels (above 90% of incident cases) does not result in the eradication of disease. In order to understand the nature of this continuing base level of incidence, the proportion of new cases caused by progression to primary disease, reinfection, or reactivation of a previous infection, is explored. Increasing the case detection rate from 80% to 100%, while keeping the cure rate constant at 80%, results in a reduction of incidence, as seen above. Increasing the case detection rate reduced the incidence of primary and reinfection cases, but left the incidence rate of reactivation cases nearly constant at 11 cases per 100,000. (Figure 9-19) Case detection combined with rapid and effective cure reduce the transmission of disease, and so the numbers of primary disease decrease soon after these are implemented. However, detection and treatment of active cases

does not reduce the incidence rates of reactivation cases, which continue to emerge from the pool of latently infected individuals.

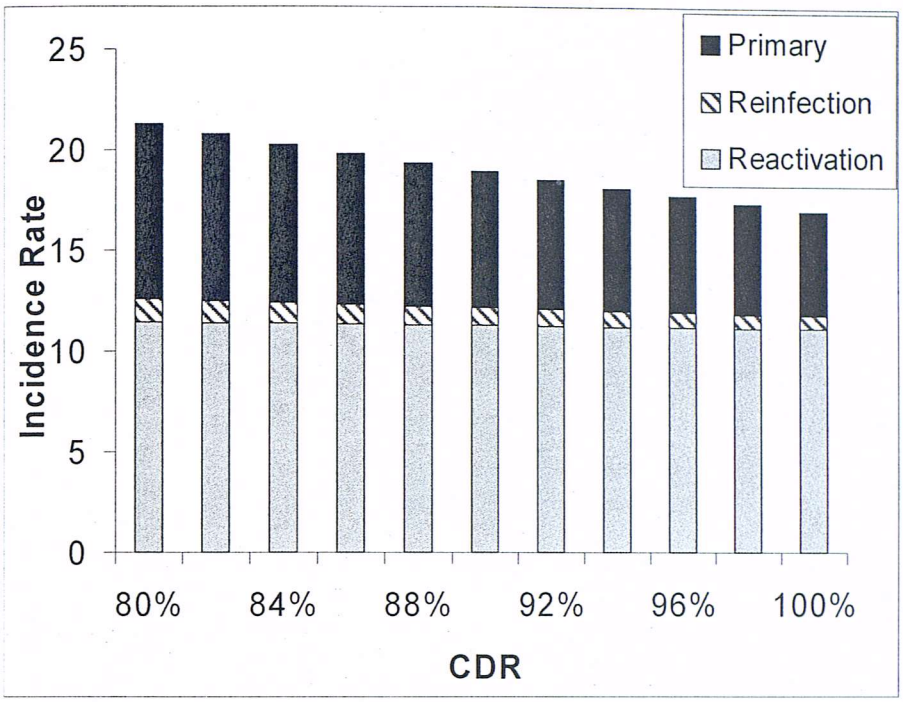


Figure 9-19. ***Increases to the case detection rate reduce the incidence of cases caused by primary disease and reinfection.*** Simulations were run as described in the text for Scenario 3. During the final ten years of the simulations, the case detection rate was set at values from 80% to 100%, while the treatment cure rate was held constant at 80%. No drug testing or resistance treatment was included. Shown is the annual incidence rate (as cases per 100,000) at the end of the simulation, by type of case. Reactivation cases are due to reactivation of an earlier infection, primary cases are due to direct progression after infection, and reinfection is due to direct progression after reinfection.

In order to reduce remaining cases due to reactivation of earlier infection, other control measures need to be implemented. This includes active screening to detect individuals with latent infection, and chemoprophylaxis treatment to reduce the rates of progression to active disease. Targeting latent infections to prevent new cases may have more impact than simply detection of new incident cases, since in this scenario ongoing transmission accounts for less of new cases than does reactivation of old infection.

In these simulations, the detection and treatment of new cases of active disease was implemented at high levels, resulting in a reduction of disease to low levels. The

remaining incidence results largely from reactivation of latent infections obtained many years earlier. The next control challenge in this scenario is reducing the final incidence of disease and seeking elimination. In order to accomplish this, active screening to detect and treat latently infected individuals is needed. In addition, the already high levels of case detection and treatment cure need to be maintained and where possible increased.

(2) Importance of DST and resistance cure

Increasing the rates of drug testing and resistance treatment reduces the incidence of resistant cases. As seen in **Figure 9-20**, higher rates of drug testing results in a reduction in the resistance incidence. Increasing the proportion of incident cases receiving drug testing and resistance treatment (pDST) from 0 to 100% results in a decrease in final resistance incidence from 3 to 1 cases per 100,000. This improvement in detection and treatment of resistance does not significantly affect the incidence of sensitive cases, which stays constant at near 18 cases per 100,000 in these simulations. (Figure 9-20)

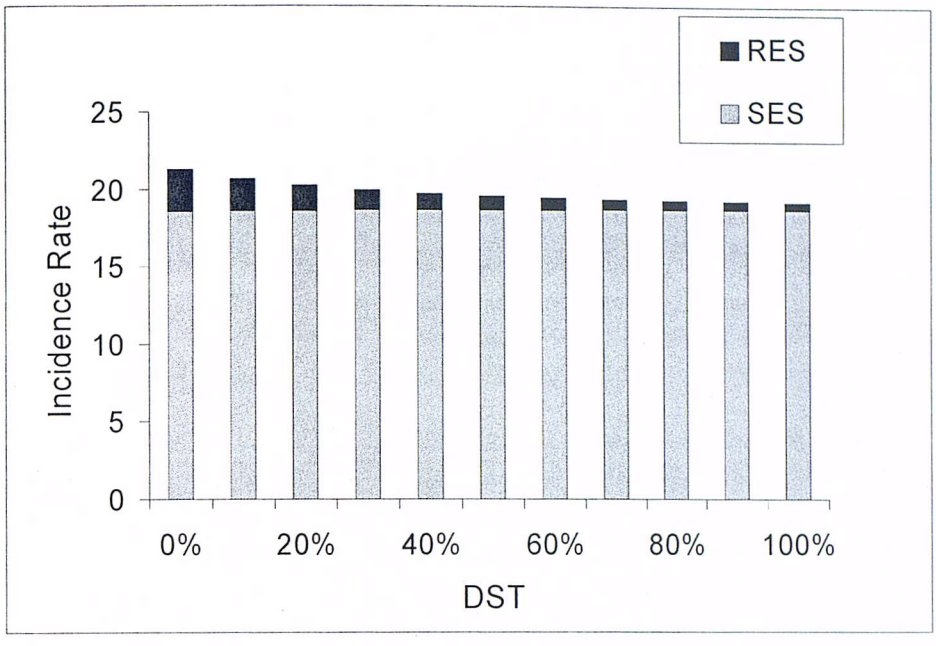


Figure 9-20. ***Increases to the drug susceptibility testing rate (DST) reduces the incidence of resistant cases.*** Simulations were run as described in the text for Scenario 3. During the final ten years of the simulations, the case detection rate was set at 80%, the cure rate for DS cases at 80%. The proportion of incident cases receiving DST and resistance treatment was set at values between 0% and 100%. The cure rate for singly resistant and MDR cases is set at 70% and 50%, respectively. Shown is the annual incidence rate (as cases per 100,000) at the end of the simulation, by resistance status.

(3) **Resistance from primary and reactivation cases**

The proportion of resistance found in primary and reactivation cases is next examined. When no drug testing or resistance treatment is offered, 90% of the drug resistance incidence is due to ongoing transmission of primary cases. The implementation of drug testing and resistance treatment reduces the incidence of resistance amongst primary cases, but does not significantly affect that amongst reactivation cases. When 100% of incident cases receive drug testing, the incidence of resistance amongst primary disease cases is reduced from 2 to 0.2 cases per 100,000. The incidence of resistance amongst reactivation cases is reduced from 0.2 to 0.1 cases per 100,000. After 10 years of 100% DST, 65% of the resistant incidence is due to ongoing primary transmission. (

Figure **9-21**)

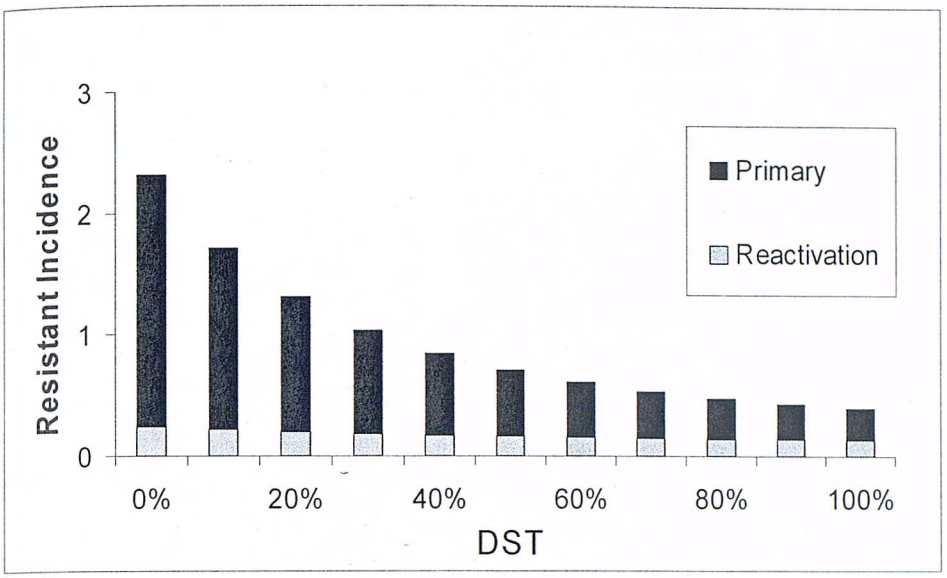


Figure 9-21. ***Increases to the DST rate reduce the incidence of resistant cases caused by primary disease.*** Simulations were run as described in the text for Scenario 3. During the final ten years of the simulations, the case detection and cure (of drug sensitive cases) rate was set at 80%. The drug testing rate of new incident cases was set at values between 0% and 100%, as shown. The cure rate for singly resistant and MDR cases is set at 70% and 50%, respectively. Shown is the annual incidence rate (as cases per 100,000) at the end of the simulation, by type of case. Reactivation cases are due to reactivation of an earlier infection, primary cases are due to direct progression after infection.

Table 9-2. Scenario 1: First-line Control Measures: Impact of improvements to control parameters on incidence in high burden country

		Incidence rate	Incidence DS cases	Incidence DR cases
Control measure		Incidence rates after 10 years this control		
CDR	Cure rate			
40%,	60%	63.6	55.2	8.4
50%,	60%	53.8	45.7	8.2
60%,	60%	46.0	38.0	7.9
70%,	60%	39.7	32.0	7.7
80%,	60%	34.7	27.2	7.5
90%,	60%	30.8	23.4	7.3

Control measure		Incidence rate	Incidence DS cases	Incidence DR cases
CDR	Cure rate	Incidence rates after 10 years this control		
40%,	80%	55.1	48.5	6.6
50%,	80%	45.6	39.3	6.3
60%,	80%	38.3	32.2	6.1
70%,	80%	32.8	26.8	5.9
80%,	80%	28.5	22.7	5.8
90%,	80%	25.2	19.5	5.7

Control measure		Incidence rate	Incidence DS cases	Incidence DR cases
CDR	Cure rate	Incidence rates after 10 years this control		
40%,	90%	51.9	45.9	5.9
50%,	90%	42.6	36.9	5.7
60%,	90%	35.6	30.1	5.5
70%,	90%	30.3	25.0	5.4
80%,	90%	26.4	21.1	5.3
90%,	90%	23.4	18.1	5.2

Scenario 1: Second-line Control Measures: Impact of improvements to control parameters on incidence

Control measure			Incidence rate	Incidence DS cases	Incidence DR cases
CDR	Cure rate	DST	Incidence rates after 10 years this control		
40%,	60%	0%	63.6	55.2	8.4
40%,	60%	50%	63.4	55.2	8.2
40%,	60%	100%	63.4	55.2	8.2
60%,	80%	0%	38.3	32.2	6.1
60%,	80%	50%	37.4	32.2	5.2
60%,	80%	100%	37.0	32.2	4.8

Table 9-3. Scenario 2: First-line Control Measures: Impact of improvements to control parameters on incidence in high burden and resistance region

Control measure		Incidence rate	Incidence DS cases	Incidence DR cases
CDR	Cure rate	Incidence rates after 10 years this control		
40%,	40%	90.2	52.4	37.8
50%,	40%	81.9	43.5	38.4
60%,	40%	75.4	36.5	38.8
70%,	40%	70.2	31.1	39.1
80%,	40%	66.1	26.8	39.3
90%,	40%	63.0	23.4	39.5

Control measure		Incidence rate	Incidence DS cases	Incidence DR cases
CDR	Cure rate	Incidence rates after 10 years this control		
40%,	60%	78.2	47.7	30.5
50%,	60%	69.6	39.0	30.6
60%,	60%	63.0	32.4	30.6
70%,	60%	58.0	27.3	30.7
80%,	60%	54.3	23.5	30.8
90%,	60%	51.4	20.5	30.9

Control measure		Incidence rate	Incidence DS cases	Incidence DR cases
CDR	Cure rate	Incidence rates after 10 years this control		
40%,	80%	69.5	44.0	25.5
50%,	80%	60.9	35.5	25.4
60%,	80%	54.6	29.3	25.4
70%,	80%	50.1	24.6	25.4
80%,	80%	46.7	21.1	25.6
90%,	80%	44.1	18.4	25.8

Scenario 2: Second-line Control Measures: Impact of improvements to control parameters on incidence

Control measure				Incidence rate	Incidence DS cases	Incidence DR cases
CDR	Cure rate	DST		Incidence rates after 10 years this control		
40%,	40%	0%		90.2	52.4	37.8
40%,	40%	50%		92.4	52.1	40.3
40%,	40%	100%		94.5	51.8	42.7
40%,	80%	0%		69.5	44.0	25.5
40%,	80%	50%		68.7	44.0	24.7
40%,	80%	100%		68.2	44.0	24.2
				90.2	52.4	37.8

Table 9-4. Scenario 3: First-line Control Measures: Impact of improvements to control parameters on incidence in low burden region

Control measure		Incidence rate	Incidence DS cases	Incidence DR cases
CDR	Cure rate	Incidence rates after 10 years this control		
80%	80%	21.3	18.6	2.7
85%	80%	19.8	17.1	2.8
90%,	80%	18.9	16.1	2.8
95%,	80%	17.7	14.9	2.8

Control measure		Incidence rate	Incidence DS cases	Incidence DR cases
CDR	Cure rate	Incidence rates after 10 years this control		
80%	90%	19.5	17.2	2.31
85%	90%	18.1	15.7	2.34
90%,	90%	17.3	14.9	2.35
95%,	90%	16.1	13.8	2.36

Control measure		Incidence rate	Incidence primary cases	Incidence reinfection cases	Incidence reactivation cases
CDR	Cure rate	Incidence rates after 10 years this control			
80%	80%	21.3	8.7	1.16	11.4
85%	80%	19.8	7.5	0.99	11.3
90%,	80%	18.9	6.8	0.88	11.3
95%,	80%	17.7	5.8	0.74	11.2

Scenario 3: Second-line Control Measures: Impact of improvements to control parameters on incidence

Control measure				Incidence rate	Incidence DS cases	Incidence DR cases
CDR	Cure rate	DST		Incidence rates after 10 years this control		
80%,	80%	0%		21.3	18.6	2.7
80%	80%	50%		19.6	18.7	0.9
80%	80%	100%		19.2	18.7	0.5

VII. Discussion:

VII. A. High Burden Countries

The WHO has classified 30 countries as high burden countries, which together account for over 90% of all TB cases globally. The incidence rates in these countries range from about 60 to over 200 cases per 100,000. (WHO 2003) (Netto, Dye et al. 1999) Only several of these countries were represented in recent drug resistance surveillance studies, allowing limited estimates of resistance prevalence amongst the HBC. Amongst the countries participating in the resistance surveillance studies, resistance prevalence ranged from 1 to 37%. Several countries reported a high incidence rate but low resistance (under 10%), including Malaysia, Nepal, Botswana, South Africa (Mpumalanga district), and Morocco (Casablanca). Other high burden countries showed drastically high resistance levels of 30-37%, including the Russian Federation, Latvia, Estonia, and China (Henan Province). (WHO/IUATLD 2000)

VII. B. Scenario 1: High incidence low resistance

In this scenario, there is a high burden of disease and a low level of drug resistance. The institution of control at the level of 40% case detection with a 60% cure rate has led to a gradual reduction in disease, while selecting for resistance. In order to continue to reduce disease in this scenario, the case detection and cure rates need to be increased. These two control parameters have the greatest influence on the course of the epidemic in this scenario. Increasing other parameters, including time to treatment, drug testing, and resistance cure, has a comparably small impact on disease levels.

This scenario has some similarities to regions with high disease and low resistance levels. These include parts of South Africa (Mpumalanga province),

Botswana, Malaysia, Nepal, Singapore, Barcelona Spain, Korea, and Guinea. (WHO/IUATLD 2000)

(1) CDR and cure rates: Limits to case detection in developing country settings

Increasing the case detection rate in a developing country setting involves a number of challenges. Barriers to increasing case detection may include health infrastructure and limited access to health care. Only individuals who have routine access to heath services are likely to be detected through passive screening, in which cases are diagnosed from amongst patients who report with symptoms. In addition, there may be inappropriate diagnosis due to lack of familiarity with recognizing the symptoms and treatment of TB. Diagnosis may also be affected by the effectiveness and use of diagnostic methods. (Shimao 1998) Furthermore, in many developing country settings, cases may be diagnosed by haphazard methods. Often practitioners, outside of regulated DOTS programs, will begin treatment after recognizing symptoms through routine physical examination. Such presumptive treatment is given without mycobacterial confirmation or chest x-ray. (Shimao 1998)

There may also be some real or program limits to case detection. Most new cases detected under DOTS programs reflect those previously detected under non-DOTS programs. As a global average, case detection rates under any program have remained near 40% of estimated incidence for nearly 20 years. (WHO 2002; WHO 2003) Programs need to address how to reach new, currently undetected, cases that may be beyond reaches of the health care systems in these countries. Increasing the case detection rate in the high burden countries will mean reaching sectors of the population currently beyond the reach of general health services.

An alternative theory is that there is a higher true case detection rate, but that current estimates of incidence are limited. First, the estimates of the proportion of cases detected depend in turn on estimates of the true incidence rate. If incidence rates have been overestimated, then the predicted undetected cases may not exist. Second, these cases may exist, and receive treatment, but not be reported to national programs. This may be of particular concern in private sector treatment removed from national regulation and surveillance. (WHO 2002; WHO 2003)

(2) Constraints to expansion

The most common constraints to expanding DOTS coverage in high burden countries have been identified as lack of technical support, health infrastructure, decentralization of health services and private sector treatment. (WHO 2003) Thus, increases to case finding in these settings should include improvements to health infrastructure, expansion of coverage of access to health care as well as NTP programs, and education of practitioners on symptoms and diagnosis of active cases.

Increasing the average treatment cure rate will similarly require a number of challenges to be met. These may include improvements to health care, NTP programs, patient management, and steady supply of quality drugs. In addition, strong training of medical officers and technical support is required. Much of this is achieved through expansion of DOTS, which provides structure for a regulated and coordinated global treatment program.

In addition, bringing cases which are usually detected and treated in the private sector into public NTP programs is important. Throughout the high burden countries, the treatment success rates are higher amongst cases treated under DOTS programs than non-DOTS. Cases treated in the private sector generally have shown lower cure rates.

Another useful measure is reporting to organized centers of excellence, which offer proficient diagnosis and treatment, as well as a high level of supervision and management. This is particularly important for cases which have failed previous courses of treatment, have suspected resistance, or are at high risk for failure.

(3) Treatment waiting time

Reducing the time to treatment, in this scenario, results in only a small reduction in incidence. This is limited by the still low case detection and cure rates. Increasing the case detection and cure rates has far greater impact on the course of the epidemic than changing the waiting time to treatment.

A number of studies have proposed introducing faster diagnostic methods, suggesting that wide scale availability of faster diagnosis would significantly improve the control of TB in HBCs. The possible benefits of large scale faster diagnosis have been suggested, but not analyzed in models of TB control. For example, using a PCR-based diagnostic test could conceivably reduce the time to diagnosis from 3+ weeks to less than 1 week. (Heymann, Brewer et al. 1997) Other means to reducing the time to treatment include improved general access to medical resources, improvements to overall health infrastructure, and education on symptoms and treatment of TB. Again, each of these measures may result in reduced average case time to treatment, thus reducing ongoing transmission and disease incidence.

VII. C. Scenario 2: High burden high resistance

This second scenario represents the most serious of public health concerns, in which a high prevalence of resistance has already emerged, and if left unchecked, may continue to increase and develop into self-sustaining cycles of a resistant epidemic. The most important concern in this scenario is first improving program conditions. Program coverage should not be expanded or wide-scale second line regimens offered,

until these conditions have improved to the extent that new drug sensitive cases are treated effectively with a low risk of developing resistance.

The largest uncertainty in model predictions comes from parameter estimates for the probability of acquiring resistance given treatment failure and the relative transmission rate of resistant strains. If the resistant strains have equal transmission to the drug sensitive, the emerging resistant epidemic will be even more difficult to control, and continuing or expanding first line treatment under current program conditions will increase selection for resistance and exacerbate the situation.

(1) Improving program conditions

Increasing the treatment cure rates and reducing the probability of acquiring resistance is likely to involve many factors outside the scope of national TB treatment programs. This may include improvements to general health care infrastructure and access. Poor socio-economic conditions have also been associated with higher rates of acquired resistance. Higher rates of resistance have also been found in regions with otherwise strong NTPs, which maintained high detection and cure rates, but which were torn by war or other social and political conflicts. Such turmoil may facilitate conditions in which optimal treatment patterns are far more difficult to obtain, and may lead to haphazard drug usage. (Chaulet and Zidouni 1998) (WHO/IUATLD 2000)

One of the most important considerations in reducing rates of acquired resistance is providing a steady supply of quality drugs for the duration of patient treatment. Without assurances of a steady supply needed to complete a course of treatment, patients may end up on a sub-optimal regimen in order to conserve drug resources, or may take drugs intermittently or only for a shortened duration. Each of these could rapidly select for resistance, particularly if cases continue on effective monotherapy. (Iseman 1999) (Chaulet and Zidouni 1998) Rates of acquired resistance

have also been correlated with program errors, including misdiagnosis, effective monotherapy, and delayed treatment. Improvements to patient supervision and care may decrease the rate of treatment errors and with this the probability of acquired resistance. (Janmeja and Raj 1998) (Mahmoudi and Iseman 1993; Chaulet and Zidouni 1998) Referring cases to specialized centers, which are able to provide a standardized high level of care, may both increase the cure rates and reduce the probability of resistance.

(2) Drug resistance treatment

Surveillance of drug resistance on a regional and individual case basis is an important consideration in this scenario. However, this may not be feasible given limitations on financial and technical resources. Drug testing requires additional technical expertise and laboratory resources. Second line regimens are also prohibitively expensive and because of longer duration, increased toxicity and side effects, these regimens are more difficult to manage. Programs that are unable to achieve high cure rates on the shorter first line SCC regimens are unlikely to offer even reasonable success rates with the second line regimens. In addition, offering second line regimens with low levels of regulation and management allows the opportunity for rapid selection of higher levels of resistance. This would select for resistance to all of the current drugs, increasing the prevalence of cases that are untreatable under these conditions. (Loddenkemper, Sagebiel et al. 2002) (Bastian, Rigouts et al. 2000) (Bahrmand, Velayati et al. 2000) (Dye, Williams et al. 2002)

Some programs have been able to include specialized MDR treatment within existing program conditions. High levels of success have been achieved in Peru, where MDR cases are referred to separate treatment centers of expertise, where cases receive high level of care and supervision. Establishing such regional centers for resistance

treatment is a plausible alternative that may prove useful in other high burden settings. (Loddenkemper, Sagebiel et al. 2002) (Bastian, Rigouts et al. 2000)

VII. D. Scenario 3: Low incidence

The third scenario presented is of a relatively low incidence region with a medium level of drug resistance, which has implemented high levels of first line control. The greatest benefits in this scenario would come from control measures not included in our model, which would involve the active detection and treatment of latently infected individuals.

Increasing the case detection rate slightly reduces disease levels. Similarly, improving the cure rate for drug sensitive cases results in a small decrease in disease levels. Even when both the case detection and cure rate are at very high levels (above 90%), there remains a continual base incidence of new drug sensitive cases. More than 90% of these incident cases were caused by reactivation of an earlier infection. Increases to case detection and case cure rate reduce the incidence of cases caused by primary progression and reinfection, but do not significantly influence the incidence of reactivation cases on the timescale of ten years examined here.

In order to reach the elimination phase of TB disease, control measures must be targeted to reduce the incidence of cases caused by reactivation of an earlier infection. Such measures include active screening for latent infection and chemoprophylaxis treatment to reduce the rate of progression to active disease. In addition, control priorities should be to maintain, and where possible increase, the already high levels of case detection and cure. Instituting drug testing of new incident cases, concomitant with effective resistance treatment, results in a decrease in resistance. However, as resistance accounted only for a small proportion of incidence, even a strong second line program will not affect the continuing incidence of drug sensitive cases.

(1) Priority setting

The priorities in this setting are to move from a focus on treatment to prevention. In order to enter the elimination phase of the TB epidemic, the base level of incidence caused by reactivation of latent infections must be reduced through active case finding and chemoprophylaxis treatment. Active screening programs are best targeted to high risk groups, of which the most significant are immigrants from high burden regions and contacts of infectious cases. (Lillebaek, Andersen et al. 2001) (Gilpin and Hammond 1987; CDC 1998; Dasgupta, Schwartzman et al. 2000; Marks, Taylor et al. 2000; Society 2000; Broekmans, Migliori et al. 2002)

These recommendations are in line with those in many of the low incidence developed countries. In Europe, the recommendations include active screening programs, particularly amongst high-risk groups. (Broekmans, Migliori et al. 2002) In the United States, recommendations for control also focus on increased active case finding and preventative treatment. This includes screening within high-risk groups such as foreign-born, contacts of infectious cases, and AIDS cases. (CDC 1998)

(2) Reducing incidence of reactivation disease

The best means to target new reactivation cases is through active screening to detect latently infected individuals and chemoprophylaxis treatment to reduce the rate of progression to active disease. Such active screening programs have been implemented in various degrees throughout much of the developed world. In particular, screening amongst contacts of infectious cases and high risk groups have been effective means of finding newly infected and disease cases.

Contact tracing, combined with chemoprophylaxis treatment for infected individuals and full treatment for new cases, has been shown to reduce disease levels. (Styblo, van Geuns et al. 1984) Models have been used to illustrate the benefits of

active screening programs. Other models have shown the cost effectiveness of contact tracing in terms of preventing new cases, which would have to undergo full medical treatment. Models have also shown the benefit, in terms disease reduction, of contact tracing and chemoprophylaxis treatment. (Murray and Salomon 1998; Murray 2002)

Cost effectiveness studies have shown the benefits of active screening programs amongst immigrants. Chemoprophylaxis treatment of immigrants from regions of high resistance reduces health costs. In addition, using regimens appropriate to these patterns, and not using INH when there is high RIF prevalence, reduces selection for MDR and concomitant increased health costs. (Khan, Muennig et al. 2002)

Difficulties in expanding case finding programs in low incidence countries may include several obstacles. First, general knowledge about TB disease and symptoms in the general population may not be sufficient. This is central to maintaining rapid case reporting, and becomes increasingly important in areas where TB is uncommon and failure to recognize symptoms may cause delays in case detection. Second, as case numbers drop, local authorities may become less familiar with detecting and treating cases, and treatment standards may fall. If necessary, regional centres may be established which maintain the technical expertise for diagnosis and treatment of cases. Local authorities and health practitioners must still be educated and alerted to recognize the symptoms of active TB and the procedures for handling cases. (American Thoracic Society 2000; Society 2000; Broekmans, Migliori et al. 2002; Blumberg, Burman et al. 2003)

(3) Relative importance of ongoing transmission

In this scenario, a high proportion of incidence was due to reactivation of latent infections. The relative importance of ongoing transmission to the continuation of the epidemic in such settings has been discussed in numerous studies. High levels of

primary disease have been found under strong control programs, as estimated by contact tracing and strain testing.

VIII. Summary of Findings

In this chapter, priorities in control of local TB epidemics are further explored. Three scenarios, with representatively different levels of disease burden and control, were examined. The impact of implementing the first and second line control measures, as described in the previous chapter, is tested in each of these settings.

The first setting is a high burden country with a low prevalence of resistance. There is a medium level of control, with a case detection rate of 40%, treatment cure rate of 50%, and no drug testing or resistance regimens. In this scenario, the control priorities lie with reducing the disease burden through increasing case detection and treatment cure rates.

The second setting is a high burden country with a high prevalence of resistance. There is a high level of case detection, but poor treatment, which has led to a growing resistance problem. In this scenario, the control priorities are two-fold: reduce disease burden through increasing case detection, and control the emerging resistance epidemic by improving program conditions and increasing treatment cure rates. The availability of resources will be a key factor in determining how control should be improved. First, program conditions should be improved ensuring high rates of treatment success with low rates of acquired resistance. Second, case detection rates should be expanded. And third, resistance regimens should be given either automatically to cases failing first line regimens, or to cases confirmed to have resistance after drug susceptibility testing.

The third setting is a low burden country, with strong first line control, entering the elimination phase of TB. The control priorities in this setting are markedly

different: with a strong control program already in place and a low incidence rate, priorities may now change to those consistent with an elimination phase. This includes improvements to all elements of the first and second line program. While high rates of case detection and cure have reduce the incidence of primary disease, a low base level of disease due to reactivation of previous infection. In order to reduce incidence towards final elimination, additional control measures are needed. These should be targeted towards detection of latently infected individuals and chemoprophylaxis treatment to reduce rates of progression to primary disease.

The two high burden settings, typically found in developing countries, generally have limited financial and technical resources. As the disease burden is still high, priorities lie with reducing disease levels through detection and treatment of cases. There is unlikely to be remaining resources available for introducing second line control in these settings; and doing so would bring only small benefits in terms of disease reduction compared to increased rates of detection. Further, introducing second line drugs under poor conditions allows for selection for resistance to more drugs, resulting in accumulation of cases increasingly difficult to treat under any program conditions. Rather, limited resources should be focused on reducing disease burden by increasing coverage and cure rates of the first line program, as much as possible.

IX. Conclusions

1. **In high burden countries priorities should lie with increasing case detection and treatment cure rates.**
2. **In high burden countries, resistance may emerge rapidly, particularly if there have been poor program conditions for a prolonged period. In these situations, the priorities lie with first improving program conditions such that new drug sensitive cases are treated effectively with a low rate of**

acquired resistance. Only once the first line program has been strengthened should DST and resistance treatment be included in order to reduce the resistance prevalence.

3. **In low incidence countries, priorities lie with eliminating disease and reducing resistance. In order to accomplish both, improved case detection (above already high levels) as well as drug susceptibility testing of all incident cases should be implemented. In addition, active screening to detect latently infected individuals and chemoprophylaxis treatment are needed to reduce incidence of reactivation cases.**

10. Conclusions:

The emergence, transmission and control of resistance within TB epidemics

In this thesis, mathematical models are used to inform on the process for the emergence, transmission, and control of resistant tuberculosis epidemics. This work contributes to the understanding of the conditions under which resistance emerges and reaches high levels, providing insight as to why there is such variability in the rates of resistance found in surveillance. The balance of priorities in control of resistance is also considered within the context of continuing epidemics and under existing program conditions. In this concluding chapter, I highlight key questions for future research, including mathematical analyses, experiments, and control.

In terms of control for overall control for epidemics and resistance, the priorities continue to lie with increased case detection and treatment, particularly in the 22 High Burden Countries. The analysis presented here should alert additional concern over control of resistance in regions with poor treatment practices and the possibility for increases in the resistance prevalence to alarming levels. Despite the range of difficulties presented to control in these regions, efforts need to be focused on improving the conditions for case detection, management, and efficacy of treatment. It is further hoped that the guidelines for control of resistance presented here will inform policy, showing the need for tuberculosis control programs in all environments to address resistance, and to assist the process of deciding how to best include the management of resistance within current programs.

I. Global burden of TB resistance

Resistance to the commonly used anti-mycobacterial drugs was first observed shortly after the introductive of effective chemotherapy in the 1950s. (Mitchison 1950; Buck 1952; Hobby 1952; Middlebrook 1952; Steenken 1952; Szybalski 1952; Barnett 1953) Studies throughout the 1960s and 1970s showed that resistance emerged rapidly under the common regimens of the day, which often involved the use of only one drug. (Mitchison 1969; Mitchison 1970) Such monotherapy reduced mortality rates, prolonging the life of patients, although a high proportion remained infectious. Such chronically infectious cases.were at high risk of developing resistance. When these limitations of monotherapy were realized, regimens began to be improved to include two drugs, and were shown to reduce the rates of acquired resistance to either of the drugs used. (Medical Research Council 1955; Crofton 1960; Barry 1964; Hobby, Johnson et al. 1969; Tripathy, Menon et al. 1969) With the introduction of rifampicin in the 1970s and the establishment of short course chemotherapy regimens involving at least four drugs, significantly higher cure rates were achieved and resistance appeared to be less of a problem. (Dickinson and Mitchison 1970; Fox and Mitchison 1975; Fox and Mitchison 1976; Mitchison 1979)

In global surveys of TB drug resistance over the past decade, resistance has been found in every region of the world surveyed. It remains unclear, however, whether resistance poses a serious problem to the control of TB epidemics. In some localized regions, the prevalence of resistance has risen to high levels, contributing to over 30% of the case incidence. In other regions, resistance remains at low levels and is largely managed by programs based on first line treatment of drug sensitive cases. (WHO/IUATLD 1997; WHO/IUATLD 2000; WHO 2003) What distinguishes these

scenarios, in terms of where resistance will emerge to high levels? And, under what conditions does resistance pose a serious threat to the control of local TB epidemics?

II. Summary of main findings

The understanding of the spread, persistence, and control of *M. tuberculosis* has been assisted at many levels by analysis based on mathematical models. Over the past decade a number of studies have used mathematical models to explore the dynamics of TB epidemics, as was reviewed in Chapter 4. In this thesis, mathematical models have been developed and used to investigate the dynamics of the emergence of resistance within TB epidemics and strategies for their control. This work contributes to the understanding of tuberculosis epidemics and the evolution of resistance on several levels. First, the model developed here offers a framework for considering the emergence and persistence of resistant TB epidemics, and insight into the conditions that favor the emergence of resistance. Second, these models have been used to consider priorities for control of resistance.

II. A. Emergence of resistance

The conditions for the emergence of resistance within TB epidemics have been examined in this thesis, using the model for the emergence and transmission of resistant TB presented in Chapter 5. The model presented here incorporates both acquired and primary resistance, as well as interaction between sensitive and resistant strains, which as outlined in Chapter 4, are important points not included in previous models. This allows for consideration of the conditions for emergence of resistance during treatment, as well as for competition with other strains and transmission through the population. On a local level, the emergence of resistance occurs through inadequate treatment that allows for the selection of resistance. These practices influence the rates of *acquired*

resistance. On a regional level, the ability of the emerging resistant strains to transmit infection, compete with other strains, and continue an epidemic will determine the success of the resistant pathogens and the levels of *primary resistance*.

As for all modeling studies, the robustness of the conclusions are limited by the quality of parameter estimates. The sensitivity analysis presented in Chapter 6 provides an understanding of how the model outcome depends on the parameter estimates, and showed that within the range of parameter uncertainty, the model can still delineate important conclusions. As expected, the model shows increasing treatment rates reduces disease and selects for resistance, dependent on the quality of treatment. Under good control, with high treatment rates and efficacy, there is a lower incidence of disease, as well as a limited emergence and spread of resistance. In comparison, under poor control there is higher incidence as well as greater range in incidence, reflecting a greater opportunity for spread. Further, the low cure and higher acquired resistance rates under poor control result in higher resistance levels and greater range in resistance prevalence. In order to address potentially serious emerging epidemics in such regions, the emergence of resistance should first be minimized by improving control practices, focusing on quality of treatment and then expanding coverage.

Examining model behavior at equilibrium brought further understanding as to the conditions under which resistant epidemics would emerge. Solving the model at equilibrium also allowed for the definition of the basic and effective reproductive numbers for each strain in terms of model parameters. This analysis provided insight into the conditions for the emergence of resistance within TB epidemics. This can best be understood by comparing the effective reproductive numbers, which reflect on the strain fitness. First, competition between sensitive and resistant strains depends upon their relative fitness. Second, resistance cannot be eradicated unless all sensitive strains are also eradicated, as resistance will continue to emerge from the treatment of sensitive

cases. Third, treatment has a differential impact on the fitness of each strain, with the implication that the conditions for eradication of each strain will differ, making it possible for one strain to succeed while another is removed under the same program. Maintaining low cure rates over a high proportion of the population may allow for the clearance of sensitive strains, while allowing resistance to increase. In order to reduce the prevalence of all strains, high notification and cure rates are required.

II. B. Control of resistance

In the final chapters of this thesis, the control practices for resistant tuberculosis were considered. In Chapter 7, a model for the control of tuberculosis is presented, based on the model for emergence and transmission developed in Chapter 4. The model accounts for control practices including case detection, drug susceptibility testing, and treatment with different regimens. The model also incorporates treatment times, including time to treatment, the duration of treatment, and time to return drug testing results.

Using this model, the control of resistance is explored in Chapter 8. Introducing treatment into a population is beneficial in terms of reducing disease levels, even if the treatment offered attains only low cure rates. Treatment also selects for drug resistance, which is more prevalent under high treatment and low cure rates. Increasing treatment rates within a population shortens case duration of infectiousness, allows for faster cure, and reduces ongoing cycles of transmission, leading to reduced incidence and prevalence of disease. However, caution is necessary if there are high rates of acquired resistance, as may occur under poor control practices. The flame of an emerging resistance epidemic may be fanned by increasing the treatment rates whilst the quality of treatment remains poor.

Introducing a second line control program, including the drug testing of a proportion of treated cases and specialized regimens, reduces resistance as well as disease levels. The success of the second line program rests upon the strength of the first line program; resistant cases can only be treated once detected, and ensuring high first line cure rates reduces the emergence of new resistant cases.

In Chapter 9, the priorities for control of resistance are further developed by considering epidemics in several representative scenarios. The first two settings represent high burden countries (HBC), differentiated based on the level of resistance and control. In HBCs with low resistance, priorities should focus on increased case detection and first line cure rates. These control measures have the greatest impact on both disease and resistance levels. HBCs with high levels of resistance, which may have poor health infrastructure, low levels of DOTS, and low cure rates, face the most serious of global TB challenges. In the absence of immediate and drastic improvements in provision of TB care, treatment, and case detection, preferably supplemented by specialized treatment for resistant cases, the situation here grows more serious. As poor treatment practices are maintained, the resistance epidemic continues to grow unchecked by 2nd line control measures. The resistance problem becomes more serious with time, as highly resistant cases are produced which are not easily treated or controlled by the current system. In this scenario, the control priorities are two-fold: reduce disease burden through increasing case detection, and control the emerging resistance epidemic by improving program conditions and increasing treatment cure rates. The availability of resources will be a key factor in determining how control should be improved. First, program conditions should be improved ensuring high rates of treatment success with low rates of acquired resistance. Second, case detection rates should be expanded. And third, resistance regimens should be

given either automatically to cases failing first line regimens, or to cases confirmed to have resistance after drug susceptibility testing.

Overall, in high burden regions the priorities lie with reducing disease levels through detection and treatment of cases. As most high burden regions are found in developing countries with limited health resources, resources should be focused on improving the quality of treatment, ensuring high cure rates, and then expanding first line treatment programs. There are unlikely to be remaining resources available for introducing second line control in these settings; and doing so would bring only small benefits in terms of disease reduction compared to increased rates of detection. Further, introducing second line drugs under poor conditions allows for selection for resistance to more drugs, resulting in accumulation of cases which are increasingly difficult to treat under any program conditions.

In those settings with low incidence, which have already established strong levels of control, priorities can move beyond the 1st and 2nd line control measures examined here. In particular, active screening for the detection and treatment of latent cases, which are a cause of a high proportion of annual incidence, has the most significant impact in reducing disease. Implementing or improving 2nd line control of resistant cases is important for reducing resistance. Reductions to treatment waiting times also reduce incidence, but again this effect is small compared to that brought about by prevention of new reactivation cases.

III. Previous work

Here, I briefly review studies within the field that have led up to or addressed similar work as that in this thesis, and consider how the work and conclusions compare.

A. Models

The relevant literature on mathematical modeling studies of tuberculosis is reviewed in Chapter 2. Several key points brought out through the modeling analyses developed in this thesis have also been made in previous modeling studies.

For example, previous modeling studies of infectious diseases have shown that resistance develops under poor treatment programs. In the models developed within this thesis, as shown in Chapters 5 and 6, it is shown that resistance develops under poor treatment conditions. This work goes further than previous in showing the dynamics for the emergence of resistance over time within TB epidemics, as well as showing how the level of acquired resistance is a reflection on the treatment state of program.

The modeling of competition between multiple strains is also a theme in this thesis, and the basis of the model developed here. Previous modeling studies have examined different types of multiple strains within an epidemiological context. For example, studies have modeled the competition between drug sensitive and resistant strains within epidemics. (Lipsitch and Levin 1997; Schrag, Perrot et al. 1997; Lenski 1998; Andersson and Levin 1999) A few studies have also considered spread of sensitive and resistant strains within a TB epidemic. (Levin, Lipsitch et al. 1997; Blower and Gerberding 1998) The current work adds to field the first comprehensive analyses of multiple sensitive and resistant strains. In particular, this is the first model to incorporate together emergence of acquired drug resistance as well as spread of primary resistance, competition with multiple sensitive and resistant strains allowing for different fitness. In addition, these dynamics are considered on a historical timescale, including not only treatment, but realistic improvements to control programs that have occurred over the past century.

The work presented in Chapters 7-9 also provides the first examination of waiting times in TB epidemics. This is of particular interest given the considered importance of waiting times in treatment, in particular the time to present to health care services, and time to diagnosis and start of treatment. Previous studies have examined waiting times in the context of other epidemics. One study has examined the influence of using different distributions for waiting times during parts of traditional (S-I-R) epidemiological models. (Lloyd 2001) One of the suggestions from this work, to use an alpha distribution to more realistically model treatment times, is also incorporated into the model developed in Chapter 7. Here, inclusion of multiple sequential compartments of infectious cases, untreated and during treatment, effectively allows the distribution of waiting time from onset of case infectiousness, until case detection and treatment, as well as duration of treatment, to be non-exponential. This is important...

B. Epidemiological studies

The recent epidemiology surveys and studies of TB are reviewed in Chapter 1. Of particular relevance to this thesis are the reports from the TB Global Resistance surveys conducted over the past decade. These have shown that resistance is found in all regions surveyed, with a high degree of variability in the prevalence of resistance by region. Some of the high burden countries showed higher resistance prevalence, as did regions with lower levels of DOTS program implemented. Yet such surveys of resistance prevalence could not address what differentiates regions which develop strikingly high levels of resistance, such as those found in a few regions with over 3-5% resistance prevalence. (Raviglione, Dye et al. 1997; WHO 1999; WHO 2003)

C. Control priorities

These epidemiological surveys of resistance, combined with annual reports of regional burden of TB, allows consideration of what control has shown effective in reducing TB resistance levels. As mentioned above, WHO resistance surveys have shown overall lower levels of resistance found under DOTS programs, suggesting that the standardized treatment program is effective at preventing or reducing resistance.

WHO studies have recommended focusing on reaching global targets for case detection and cure rates. These studies prioritize the detection of new cases and bringing them under standardized treatment programs as DOTS, and suggest that these are the primary impediments to control of TB epidemics worldwide.

In some local area epidemics, there have been examples under which outbreaks of MDR-TB have been shown to be effectively controlled through improvements to the control practices and first-line treatment, without widespread second line treatment programs. For example, during the NYC MDR outbreak in the 1990s, improvements to the first line treatment program was shown to rapidly reduce and control the epidemic. (Eltringham and Drobniewski 1998; Bleed, Dye et al. 2000; Dye, Fengzeng et al. 2000; Espinal, Kim et al. 2000; Espinal, Laszlo et al. 2001; Raviglione, Gupta et al. 2001) (Alland, Kalkut et al. 1994; Friedman, Stoeckle et al. 1995; Bifani PJ 1996; Munsiff, Bassoff et al. 2002)

Other studies have examined the effectiveness of introducing standardized second-line treatment programs, such as DOTS+, within existing control programs. The WHO currently recommends introducing such DOTS+ programs in areas which have already achieved high levels of control under DOTS program, maintaining target case detection and first line cure rates. (WHO 1997; WHO 1999)

These recommendations for control priorities are similar and in agreement with those emerging from the work in this thesis. It is clear from the modeling studies

developed here, that the most important consideration in terms of controlling disease levels is the detection and effective treatment of new cases. This was shown most clearly in Chapter 6, with sensitivity analyses that showed the case detection and then the cure rates had the biggest effect on the disease as well as resistance levels. This was shown in more detail in Chapter 9, that within each of three TB epidemics varying in disease and resistance levels, increasing the case detection and cure rates were the most important considerations in reducing disease and resistance levels.

The work developed in this thesis also adds a few additional points to help understand the dynamics of control of resistance within TB epidemics. For example, it was shown in Chapter 7 that introducing second line control, with the first line program still has suboptimal case detection and cure rates, can lead to a rapid exacerbation of resistance. This suggests how introducing second line control (such as a DOTS+ program) before the basic DOTS program implemented detection and effective treatment of a high proportion of new drug sensitive cases, can lead to increases in resistance prevalence, as well as amplification of resistance due to mistreatment. This suggests, again in agreement with current WHO guidelines, that DOTS+ programs not be introduced in areas which have not met the DOTS targets for case detection and cure rates.

IV. Future directions: Experimental priorities

In this section, the experimental priorities in tuberculosis research are considered. As examined in Chapter 6, the model predictions are sensitive to uncertainty in estimating key parameters. In particular, the resistance prevalence depends heavily on the variation in estimates of the relative fitness of the resistance strain, and the probability of acquiring resistance during treatment.

For progress in the field of mathematical models for tuberculosis, refining key estimates of resistance transmission and the probability of acquiring resistance during treatment are central. In addition, the next generation of models should be expanded to include aspects of network based models, HIV coinfection, and within host properties of mycobacterial infection.

A. Resistance transmission and fitness

Strain fitness, as defined in this thesis, includes the probability of transmission of infection to a susceptible host given contact (defined as the effective contact rate) and the probability of progression to active disease given infection (known as strain virulence). Both transmission of infection and strain virulence are components of the overall strain fitness, as measured by the basic reproductive number defined here. Measurement of mycobacterial strain fitness is more complicated than providing estimates from mathematical models. As shown in Chapter 6, the relative fitness of resistant strains is of great significance in model predictions for the dynamics of resistance within epidemics. The variance in the resistant transmission rate was a primary cause of model uncertainty in resistance prevalence, in particular under poor control.

However, the relative transmission rate of resistant, and in particular MDR, strains remains a source of significant uncertainty. Experiments have suggested a wide range of transmission rates for resistant strains— from strains with reduced fitness to 'super fit' MDR strains with greater transmissibility than drug sensitive strains.

Strain fitness can be measured through contact tracing studies. One approach is to examine the proportions of contacts of sensitive and resistant cases which convert and develop active disease. This can then be compared to studies estimating the same for contacts of sensitive cases. A number of such studies have been performed, largely

in low incidence developed country settings, as reviewed in Chapter 4. Studies have also investigated the proportion of resistant cases due to ongoing transmission. This can be estimated as the prevalence of resistance amongst new and previously treated cases, assuming that new cases are caused by transmission of a resistant strain, while acquired resistance during treatment is found amongst previously treated strains.

More recently, convention has led towards using molecular epidemiological studies to examine transmission. In these studies, clustering of strains with similar genetic markers is used as a surrogate for recent transmission, and such analysis has been used to estimate the relative proportion of resistant strains in a population due to ongoing transmission. In an area of high incidence (Capetown, South Africa), the proportion of resistance due to primary transmission, as detected by RFLP strain typing, was high; over 50% of MDR cases were clustered. (Van Rie, Warren et al. 2000) In Denmark, over 70% of resistant strains were found to be clustered. (Thomsen, Bauer et al. 2000) There have also been reports of 'super fit' resistant strains which have been found more frequently in clusters and are believed to have greater transmission. (Agerton, Valway et al. 1999; Kruuner, Hoffner et al. 2001; Toungoussova, Sandven et al. 2002)

Interventions can also be designed to specifically or preferentially target resistant cases in order to reduce their contact rates, thus reducing the overall resistance fitness. Where available, resources for active case detection can be focused on diagnosed MDR cases, finding and treating contacts of these cases. This can be implemented by sending MDR cases for treatment at specialized regional centres of excellence. (Farmer 2001; Mitnick, Bayona et al. 2003) Finding and testing contacts of MDR cases can identify new latent infections or cases of disease for rapid treatment. (Loddenkemper, Sagebiel et al. 2002)

B. Acquired resistance rate

As described in Chapter 4, there is significant uncertainty in estimating the probability of acquiring resistance during treatment. The sensitivity analysis has shown that the model predictions for resistance prevalence are very sensitive to the uncertainty in the acquired resistance rate. This is most significant under poor control, when there is the potential for the emergence of a high level of resistance. Under good control, the uncertainty in estimates for the acquired resistance rate does have significant impact on model predictions for resistance prevalence, but high case detection and cure rates limit the spread of resistance.

Studies have suggested that acquired resistance accounts for only a small proportion of treatment failures, and that cases are more likely to fail treatment for other reasons. One method of estimating from surveillance has suggested that resistance arises amongst treatment failures at a rate of 13% for isoniazid, 8% for rifampicin, and 7% for MDR. (Dye and Espinal 2001) In this thesis, using the model presented in Chapter 5, the rate of acquiring resistance given treatment failure was estimated at approximately 5-15%, and depended upon the control program and disease levels. These estimates of the proportion of acquired resistance have been made assuming that there is a constant rate of acquiring resistance during treatment. However, there is also evidence that this statistic should vary with the quality of treatment. Under good control measures, there is a low rate of acquired resistance. When the control is poor, higher levels of resistance are found, particularly when there are lower rates of DOTS implementation. (WHO/IUATLD 2000)

For example, the probability of acquiring resistance has been correlated with errors in treatment, case management, and administration. (Mahmoudi and Iseman 1993) Poor treatment practices correlated to increased resistance rates include adding one drug to a failing regimen, using an inadequate regimen, not diagnosing initial

resistance, and poor patient adherence. (Iseman 1999) Use of supervised standardized treatment and a regulated drug supply has been shown to reduce the proportion of cases developing resistance. (Weis, Slocum et al. 1994) Under fully supervised and regulated DOTS programs, a lower proportion of treatment failures are seen to acquire resistance. (Weis, Slocum et al. 1994; Bastian, Rigouts et al. 2000; Dye, Williams et al. 2002) High rates of acquired resistance are seen in environments with poor control practices, less regulation, and other factors which might interfere with regular drug taking, such as poverty or war. (Iseman 1999; Loddenkemper, Sagebiel et al. 2002) Improving these control practices can not only improve the treatment cure rates, but also reduce the probability of acquiring resistance.

Experiments focused on the within host dynamics of a mycobacterial infection are crucial to unveiling the mysteries associated with acquired resistance. Useful studies include observing the selection for resistance within a patient through a timecourse of treatment. The means to undertake such experiments may be within reach; already, the use of PCR tests for resistance have allowed improvements in quantification of the relative proportion of resistant mycobacteria present in sputum. Such studies could begin to address the questions of what treatment conditions allow for the selection of acquired resistance.

V. Future directions: Implications for tuberculosis control

Tuberculosis remains a leading public health concern. With over one third of the world's population infected, an annual incidence of 9 million new cases, and nearly 3 million deaths to tuberculosis each year, tuberculosis continues to pose a health emergency. (Dye, Scheele et al. 1999; WHO 2000; Mathers, Stein et al. 2002; WHO 2003) There is still much work to bring tuberculosis epidemics under control. This

includes several frontiers, including increasing coverage of DOTS programs and case detection rates, improving and maintaining quality of treatment, and managing and treating resistance.

The first, and most important, next step is increasing coverage of current programs. An estimated 60% of annual cases remain undetected. The greatest number of cases is found in developing countries, with over 80% occurring in the 22 high burden countries (HBC). Global control efforts should focus on increasing and improving programs in these high burden areas. These efforts need to be centered around new means for detecting cases which otherwise would remain untreated. In particular, cases which occur amongst high risk groups and parts of the population which have reduced access to health services.

Second, global control priorities should focus on improving overall treatment conditions to ensure high level of cure. DOTS offers manageable and cost-effective programs which have been repeatedly shown to achieve high cure rates even under difficult conditions. Increasing the reach of DOTS programs provides improved conditions for patient care and treatment. This in turn leads to better patient outcomes, including higher cure rates and lower rates of acquired resistance. These efforts may involve the introduction of standardized DOTS programs into new regions, as well as the extension of DOTS coverage to parts of populations currently seeking private care.

Third, the problem of emerging resistance must be handled proactively. Knowing that resistance emerges under every treatment program, strategies for control of these resistance cases should be incorporated into program strategies. These strategies should be fitting to the local environment, based on an understanding of the state of the local epidemic, the control practices, and the likelihood for resistance. The analysis presented in this thesis can provide initial guidelines for setting priorities.

Of pressing concern is the treatment of resistance in those high burden countries in which there has already emerged a high prevalence of resistance. This includes regions such as the Russian Federation, Latvia, Estonia, and China. These high burden regions each show drastically high levels of resistance, of over 30%, in recent surveillance. (WHO/IUATLD 2000) Unfortunately, control in these areas may also be the most difficult to improve. Due to consistently poor treatment practices, resistance has developed to high levels in this area. In many of these areas, there are severely limited resources for health care and often only a fraction of the population has routine access to these services.

In regions with a high burden of disease and a low prevalence of resistance, concentrating on increasing coverage and quality of treatment under first line DOTS programs should be the first priority. These measures should also manage resistance. However, particular caution should be taken to ensure overall high cure rates and quality of care before expanding coverage. This should include measures for surveillance of treatment, patient outcomes, and resistance. In addition, continual surveillance for resistance will allow for rapid alerts if there are local or regional increases in resistance that could warrant changes in treatment protocols. Countries which have reported a high incidence rate but low resistance (under 10%) include parts of South Africa (Mpumalanga province), Botswana, Malaysia, Nepal, Singapore, Spain, Korea, and Guinea. (WHO/IUATLD 2000)

APPENDIX

11. Appendix 1

Model for Chapter 5

Figure 1: Model Equations

Figure 11-1. The partial differential equations representing the three-strain model presented in Chapter 5. This model includes a drug sensitive (S), a singly drug resistant (R), and a multiply drug resistant (M) strain.

$$\frac{dX}{dt} = \Pi - \lambda \quad X - \mu \; X$$

$$\frac{dL_S}{dt} = (1-p)\,\lambda_S \; X - (p\,\gamma\,\lambda + v + \mu)\,L_S$$

$$\frac{dL_R}{dt} = (1-p)\,\lambda_R \; X - (p\,\gamma\,\lambda + v + \mu)\,L_R$$

$$\frac{dL_M}{dt} = (1-p)\,\lambda_M \; X - (p\,\gamma\,\lambda + v + \mu)\,L_M$$

$$\frac{dI_S}{dt} = i\ p\lambda_S\ X + i\ v\ L_S - \ n\ (\phi_S + r\ (1-\phi_S))\ \ I_S\ \ + i\ \gamma\ p\ \lambda_S\ (L+T) - (\kappa+\mu+\mu_T)\ I_S + w_S\ T_S$$

$$\frac{dI_R}{dt} = i\ p\lambda_R X + i\ v\ L_R + r\ n\ (1-\phi_S)\ I_S - n(\phi_R + r\ (1-\ \phi_R))\ I_R + i\ \gamma\ p\ \lambda_R\ (L+T) - (\kappa+\mu+\mu_T)\ I_R + w_R\ T_R$$

$$\frac{dI_M}{dt} = i\ p\lambda_M X + i\ v L_M + r\ n(1-\phi_R) I_R - \ n\ \phi_M\ I_M + i\gamma\ p\lambda_M (L+T) - (\kappa+\mu+\mu_T) I_M + w_M\ T_M \qquad \frac{dT_S}{dt} = [\kappa + n\phi_S\ ]\ I_S - (p\ \gamma\ \lambda + \mu + w_S)\ T_S$$

$$\frac{dT_R}{dt} = [\kappa + n\phi_R\ ]\ I_R - (p\ \gamma\ \lambda + \mu + w_R)\ \ T_R$$

$$\frac{dT_M}{dt} = [\kappa + n\phi_M\ ]\ I_M - (p\ \gamma\ \lambda + \mu + w_M)\ \ T_M$$

$$\frac{dNI}{dt} = (1-i)\ p\ \lambda\ X + \ (1-i)\ \gamma\ p\ \ \lambda\ \ (L+T) + (1-i) v\ L - (\kappa+\mu)\ NI$$

$$\lambda_S = c_S\ \frac{I_S}{N} \quad , \quad \lambda_R = c_R\ \frac{I_R}{N} \quad , \quad \lambda_S = c_M\ \frac{I_M}{N}$$

$$\lambda = \ \lambda_S + \lambda_R + \lambda_M$$

$$L = L_S + L_R + L_M$$
$$I = I_S + I_R + I_M$$
$$T = T_S + T_R + T_M$$
$$N = X + L + I + T$$
$$\Pi = \mu N + \mu_T I + (\mu_T + \kappa) NI$$

Figure 2: Model flow chart

Figure 11-2. The three-strain model for the emergence and transmission of drug resistant and MDR tuberculosis is represented in this diagram. The equations for this model are given in Appendix 1. Individuals are born into the population (kept at a constant size) as susceptibles (X), and are subject to infection by the drug sensitive (S), singly drug resistant (R), or multiply drug resistant (M) strain. Infected individuals either progress directly to infectious (I) or non-infectious (NI) disease, or enter a latent state from which there is a slow reactivation rate. Infectious cases receive drug treatment and either recover (T), fail treatment and remain drug sensitive, or fail treatment and acquire drug resistance. Individuals previously infected with TB (L and T classes) may be reinfected with any of the three strains (for simplicity, lines representing reinfection are not shown here).

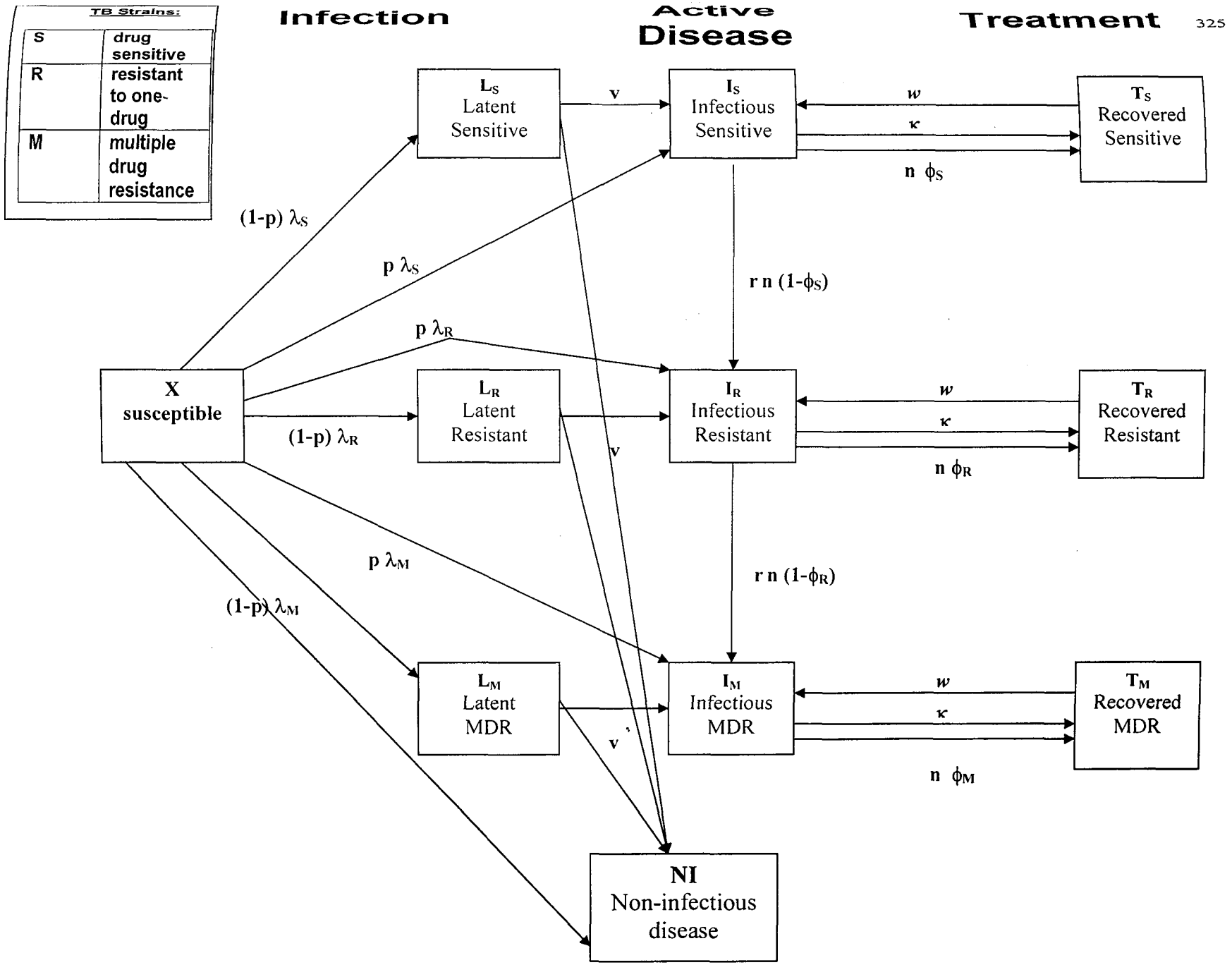

TB Strains:
S drug sensitive
R resistant to one-drug
M multiple drug resistance
Infection
Active Disease
Treatment
X susceptible
L_S Latent Sensitive
L_R Latent Resistant
L_M Latent MDR
I_S Infectious Sensitive
I_R Infectious Resistant
I_M Infectious MDR
T_S Recovered Sensitive
T_R Recovered Resistant
T_M Recovered MDR
NI Non-infectious disease
(1-p) λ_S
p λ_S
p λ_R
(1-p) λ_R
p λ_M
(1-p) λ_M
v
v
v
w
κ
n ϕ_S
r n (1-ϕ_S)
w
κ
n ϕ_R
r n (1-ϕ_R)
w
κ
n ϕ_M

Figure 3: Parameter values

Parameter	Explanation	Estimated values	Source
λ	*Per capita rate of infection* $= c\ I/N$		
c	*Effective contact rate*	5-15 (average 10) infections/ infectious case - year	(Styblo, 1991), (Blower, McLean et al. 1995), (Porco and Blower 1998), (Dye and Williams 2000; Dye and Espinal 2001)
i	*Proportion infectious*	0.4 – 0.6	(Fishman and Elias 1998) (Davies 1998)
p	*Proportion infected progress to primary disease*	5-15%	(Sutherland, Svandova et al. 1982; Sutherland, Bleiker et al. 1983) (Vynnycky and Fine 1997)
v	*Rate latent infected progress to infectious disease*	0.03%	(Sutherland, Svandova et al. 1982; Sutherland, Bleiker et al. 1983), (Vynnycky and Fine 1997)
κ	*Self-cure rate for DS cases*	10%	(Chaulet and Zidouni 1998) (Enarson and Hopewell 1998), Estimates in this chapter.
n	*Notification rate for sputum smear positive cases*		
ϕ_S	*treatment cure rate of DS cases*	*Developed: 70-95%, developing: 50-75%*	(WHO/IUATLD 2000; WHO 2003), (Chaulet and Zidouni 1998)
ϕ_R	*Treatment cure rate of single DR cases*	*Developed: 70-95%, developing: 50-75% (similar to DS cure rates)*	(Espinal, Kim et al. 2000)
ϕ_M	*Treatment cure rate of multiple DR cases*	*developed: 50-80% developing: 4-60% (reduced cure rates)*	(Espinal, Kim et al. 2000)
r	*Prob. of acquiring resistance mutation during drug treatment of DS case*	*Developed: low(5%) – high(10%) Developing: high (10-15%)*	(Chaulet and Zidouni 1998), (Dye and Espinal 2001)
μ	*Mortality rate (not TB)*	0.015	Average life expectancy 80 years
μ_T	*Mortality rate due to TB*	15-30 (mean 20)%	(Grzybowski 1991; Chaulet and Zidouni 1998; Davies 1998; Enarson and Hopewell 1998), Estimates in this chapter.

w	*relapse rate*	2-5%	(Chaulet and Zidouni 1998)

Figure 11-3. *Estimates of values for model parameters.* Unless otherwise indicated, parameter values are given as rate per year. For control parameters (including the notification, cure, and acquired resistance rates) ranges of values found in developed and developing countries are given.

Figure 4: Two strain model

Figure 11-4. The partial differential equations representing the two strain version of the model presented in Chapter 5. This model includes one drug sensitive (S) and one drug resistant (R) strain.

$$\frac{dX}{dt} = \Pi - \lambda\ X - \mu\ X$$

$$\frac{dL_S}{dt} = (1-p)\,\lambda_S\ X - (p\,\gamma\,\lambda + v + \mu)\,L_S$$

$$\frac{dL_R}{dt} = (1-p)\,\lambda_R\ X - (p\,\gamma\,\lambda + v + \mu)\,L_R$$

$$\frac{dI_S}{dt} = i\ p\lambda_S\ X + i\,v\,L_S - (n\,(\phi_S + r\ (1-\phi_S))\,)\,I_S\ + i\,\gamma\ p\ \lambda_S\,(L+T) - (\kappa + \mu + \mu_T)\,I_S + w_S\ T_S$$

$$\frac{dI_R}{dt} = i\ p\lambda_R X + i\,v\,L_R + i\,\gamma\ p\ \lambda_R\,(L+T) + r\,n\,(1-\phi_S)\,I_S - (n\phi_R + \kappa + \mu + \mu_T)\,I_R + w_R\ T_R$$

$$\frac{dT_S}{dt} = [\kappa + n\phi_S]\,I_S - (p\gamma\,\lambda + \mu + w_S)\ T_S$$

$$\frac{dT_R}{dt} = [\kappa + n\phi_R]\,I_R - (p\gamma\,\lambda + \mu + w_R)\ T_R$$

$$\frac{dNI}{dt} = (1-i)\ p\ \lambda\ X + (1-i)\,\gamma\ p\ \lambda\ (L+T) + (1-i)v\,L - (\kappa + \mu)\,NI$$

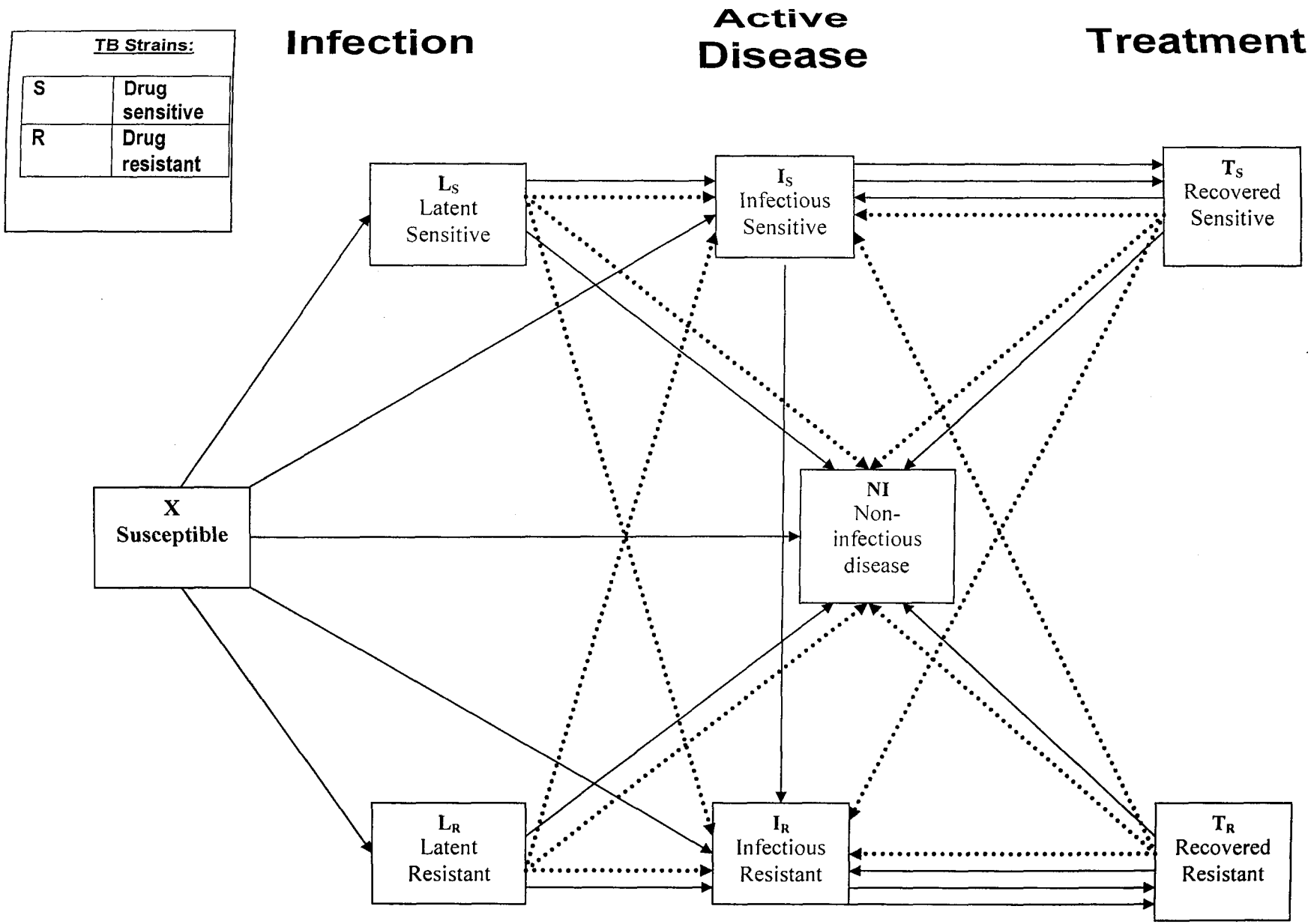
$$\lambda_S = c_S \frac{I_S}{N} \quad , \quad \lambda_R = c_R \frac{I_R}{N} \quad , \quad \lambda = \lambda_S + \lambda_R$$

$$\begin{aligned} L &= L_S + L_R \\ I &= I_S + I_R \\ T &= T_S + T_R \\ N &= X + L + I + T \end{aligned}$$

$$\Pi = \mu N + \mu_T I + (\mu_T + \kappa) NI$$

Figure 5: Two strain model flow chart

Figure 11-5. The two-strain model for the emergence and transmission of drug resistant tuberculosis is represented in this diagram. The equations for this model are given in Appendix 3. Individuals are born into the population (kept at a constant size) as susceptibles (X), and are subject to infection by either the drug sensitive (S) or drug resistant (R) strain. Infected individuals either progress directly to infectious (I) or non-infectious (NI) disease, or enter a later state from which there is a slow reactivation rate. Infectious cases receive drug treatment and are either cured (T), fail treatment and remain drug sensitive, or fail treatment and acquire drug resistance. Individuals previously infected with TB may be reinfected with either strain (reinfection shown in dotted lines).


TB Strains:
S
Drug sensitive
R
Drug resistant
Infection
Active Disease
Treatment
L_S Latent Sensitive
I_S Infectious Sensitive
T_S Recovered Sensitive
X Susceptible
NI Non-infectious disease
L_R Latent Resistant
I_R Infectious Resistant
T_R Recovered Resistant

12. Appendix 2

Model for Chapter 7

Model equations

$$\frac{dX}{dt} = \Pi - (\lambda_S + \lambda_R + \lambda_M)\, X - \mu\ X$$

$$\frac{dL_S}{dt} = (1-p)\,\lambda_S\ X - (\gamma\ \lambda + v + \mu)\, L_S$$

$$\frac{dL_R}{dt} = (1-p)\,\lambda_R\ X - (\gamma\ \lambda + v + \mu)\, L_R$$

$$\frac{dL_M}{dt} = (1-p)\,\lambda_M\ X - (\gamma\ \lambda + v + \mu)\, L_M$$

Force of infection, by strain and overall:

$$\lambda_S = c_S \frac{I_S}{N} \quad , \quad \lambda_R = c_R \frac{I_R}{N} \quad , \quad \lambda_S = c_M \frac{I_M}{N}$$

$$\lambda = \lambda_S + \lambda_R + \lambda_M$$

New active non-infectious TB cases:

$$\frac{dNI}{dt} = (1-i)\ p\ \lambda\ X + \ (1-i)\ \gamma\ p\ \ \lambda\ \ (L+C) + (1-i)v\ L - (\kappa+\mu)\ NI$$

New incident cases of active infectious TB cases, by strain type:

$$\frac{dI_S}{dt} = i\ p\lambda_S\ X + i\ v\ L_S\ \ + i\ \gamma\ p\ \lambda_S\ (L+C) - (t_1+\kappa+\mu+\mu_T)\ I_S$$

$$\frac{dI_R}{dt} = i\ p\lambda_R X + i\ v\ L_R - (\mu\ + \mu_T + n(\phi_R + r(1-\ \phi_R)))\ I_R + i\ \gamma\ p\ \lambda_S\ (L+C) - (t_1+\kappa+\mu+\mu_T)\ I_R$$

$$\frac{dI_M}{dt} = i\ p\lambda_M\ X + i\ v L_M + r_2 n(1-\phi_R)\ I_R - (\mu + \mu_T + n(\phi_M + r(1-\phi_M)))I_M + i\ \gamma\ p\lambda_S\ (L+C) - (t_1+\kappa+\mu+\mu_T)\ I_M$$

Undetected active infectious cases, by strain type:

$$\frac{dU_S}{dt} = t_1\ CDR\ \ I_S\ \ - (\mu + \mu_T + \kappa)\ U_S$$

$$\frac{dU_R}{dt} = t_1\ CDR\ \ I_R\ \ - (\mu + \mu_T + \kappa)\ U_R$$

$$\frac{dU_M}{dt} = t_1\ CDR\ \ I_M\ \ -(\mu + \mu_T + \kappa)\, U_M$$

New active infectious TB cases waiting for case detection after time tCD (for bookkeeping convenience), by strain type:

$$\frac{d\, I_S^{CD}}{dt} = t_1\ CDR\ \ I_S - (t_{CD} + \kappa + \mu + \mu_T)\ \ I_S^{CD}$$

$$\frac{d\, I_R^{CD}}{dt} = t_1\ CDR\ \ I_R\ \ - (t_{CD} + \kappa + \mu + \mu_T)\, I_R^{CD}$$

$$\frac{d\, I_M^{CD}}{dt} = t_1\ CDR\ \ I_S - (t_{CD} + \kappa + \mu + \mu_T)\ \ I_M^{CD}$$

Treated drug sensitive cases, undergo treatment course for phases 1 – 4, and either remain infectious (I) or become non-infectious (NI):

Infectious (I) DS cases (S) on treatment regimen a (a), by treatment phase (1, 2, 3, 4):

$$\frac{dT_{Sa}^{I}}{dt}[\mathrm{I}] = t_{CD}\ I_S^{CD} - (t_{ph1} + \mu + \mu_T + \kappa)\ \ T_{Sa}^{I}[\mathrm{I}] + t_{ph5}\, P_S$$

$$\frac{dT^{I}_{Sa}}{dt}[2]=(1-\theta_{Sa})\,t_{ph}\;T^{I}_{Sa}[1]-(t_{ph2}+\mu+\mu_{T}+\kappa)\;T^{I}_{Sa}[2]$$

$$\frac{dT^{I}_{Sa}}{dt}[3]=(1-r_{1})\,t_{ph2}\;T^{I}_{Sa}[2]-(t_{ph3}+\mu+\mu_{T}+\kappa)\;T^{I}_{Sa}[3]$$

$$\frac{dT^{I}_{Sa}}{dt}[4]=t_{ph3}\;T^{I}_{Sa}[3]-(t_{ph4}+\mu+\mu_{T}+\kappa)\;T^{I}_{Sa}[4]$$

Non-infectious (NI) DS cases (S) on treatment regimen a (a), by treatment phase (1, 2, 3, 4):

$$\frac{dT^{NI}_{Sa}}{dt}[1]=0$$

$$\frac{dT^{NI}_{Sa}}{dt}[2]=\theta_{Sa}\,t_{ph}\;T^{I}_{Sa}[1]-(t_{ph2}+\mu+\mu_{T}+\kappa)\;T^{NI}_{Sa}[2]$$

$$\frac{dT^{NI}_{Sa}}{dt}[3]=(1-r_{2})\,t_{ph2}\;T^{NI}_{Sa}[2]-(t_{ph3}+\mu+\mu_{T}+\kappa)\;T^{NI}_{Sa}[3]$$

$$\frac{dT^{NI}_{Sa}}{dt}[4]=t_{ph3}\;T^{NI}_{Sa}[3]-(t_{ph4}+\mu+\mu_{T}+\kappa)\;T^{NI}_{Sa}[4]$$

DS cases which are either cured (C) or have failed treatment (P, previously treated):

$$\frac{dC_S}{dt} = t_{ph4}\left[(1-w_{Sa}^{NI})\,T_{Sa}^{NI}[4] + (1-w_{Sa}^{I})\,T_{Sa}^{I}[4]\right] + \kappa(I_S^{CD} + T_{Sa}^{I}[1.4] + T_{Sa}^{NI}[1.4] + I_S + U_S + P_S) - (\mu + \gamma p\lambda)C_S$$

$$\frac{dP_S}{dt} = t_{ph4}\left[w_{Sa}^{NI}\,T_{Sa}^{NI}[4] + w_{Sa}^{I}\,T_{Sa}^{I}[4]\right] - (t_{ph5} + \mu + \mu_T + \kappa)P_S$$

Singly drug resistant cases (DR), treated with regimen A, continue infectious:

$$\frac{dT_{Ra}^{I}}{dt}[1] = t_{CD}(1-p_{DST})p_{incR}I_R^{CD} - (t_{ph1} + \mu + \mu_T + \kappa)\,T_{Ra}^{I}[1] + t_{ph5}(1-p_{DST})p_{incR}P_R$$

$$\frac{dT_{Ra}^{I}}{dt}[2] = (1-r_{MDR})t_{ph1}\,T_{Ra}^{I}[1] - (t_{ph2} + \mu + \mu_T + \kappa)\,T_{Ra}^{I}[2]$$

$$\frac{dT_{Ra}^{I}}{dt}[3] = (1-r_{MDR})\,t_{ph2}\,T_{Ra}^{I}[2] - (t_{ph3} + \mu + \mu_T + \kappa)\,T_{Ra}^{I}[3]$$

$$\frac{dT_{Sa}^{I}}{dt}[4] = (1-r_{MDR})t_{ph3}\,T_{Ra}^{I}[3] - (t_{ph4} + \mu + \mu_T + \kappa)\,T_{Ra}^{I}[4]$$

Singly drug resistant cases (DR), treated with regimen B, continue infectious and fail treatment:

$$\frac{dT^{I}_{Rb}}{dt}[1] = t_{CD}\, p_{DST}\, p_{incR}\, I^{CD}_{R} - (t_{DST} + \mu + \mu_T + \kappa)\; T^{I}_{Rb}[1] + t_{pI5}\, p_{DST}\, p_{incR}\, P_R$$

$$\frac{dT^{I}_{Rb}}{dt}[2] = (1 - r_{MDR})\, t_{DST}\, T^{I}_{Rb}[1] - (t_{pI1} + \mu + \mu_T + \kappa)\; T^{I}_{Rb}[2]$$

$$\frac{dT^{I}_{Rb}}{dt}[3] = (1 - r_1)\; \theta_{Rb}\, t_{pI1}\; T^{I}_{Rb}[2] - (t_{pI3} + \mu + \mu_T + \kappa)\; T^{I}_{Rb}[3]$$

$$\frac{dT^{I}_{Rb}}{dt}[4] = t_{pI2}\; T^{I}_{Rb}[3] - (t_{pI3} + \mu + \mu_T + \kappa)\; T^{I}_{Rb}[4]$$

$$\frac{dT^{I}_{Rb}}{dt}[5] = t_{pI3}\; T^{I}_{Rb}[4] - (t_{pI4} + \mu + \mu_T + \kappa)\; T^{I}_{Rb}[5]$$

Singly drug resistant cases (DR), treated with regimen B, become non-infectious through effective treatment:

$$\frac{dT^{NI}_{Rb}}{dt}[1] = 0$$

$$\frac{dT_{Rb}^{NI}}{dt}[2] = (1-\theta_{Rb})\, t_{ph1}\, T_{Rb}^{I}[2] - (t_{ph2}+\mu+\mu_T+\kappa)\, T_{Rb}^{NI}[2]$$

$$\frac{dT_{Rb}^{NI}}{dt}[3] = t_{ph2}\, T_{Rb}^{NI}[2] - (t_{ph3}+\mu+\mu_T+\kappa)\, T_{Rb}^{NI}[3]$$

$$\frac{dT_{Rb}^{NI}}{dt}[4] = t_{ph3}\, T_{Rb}^{NI}[3] - (t_{ph4}+\mu+\mu_T+\kappa)\, T_{Rb}^{NI}[4]$$

DR cases which are either cured (C) or have failed treatment (P, previously treated):

$$\frac{dC_R}{dt} = t_{ph4}\left[\, (1-w_{Rb}^{I})\, T_{Rb}^{I}[4] + (1-w_{Rb}^{NI})\, T_{Rb}^{NI}[4] \,\right] + \kappa(I_R^{CD} + T_{Ra}^{I}[1.4] + T_{Rb}^{I}[1.5] + T_{Rb}^{NI}[1.4] + I_R + U_R + P_R) - (\mu+\gamma p\lambda)\, C_R$$

$$\frac{dP_R}{dt} = t_{ph4}\left[T_{Ra}^{I}[4] + w_{Rb}^{I}\, T_{Rb}^{I}[4] + w_{Rb}^{NI}\, T_{Rb}^{NI}[4]\right] - (t_{ph5}+\mu+\mu_T+\kappa)\, P_R$$

Multiple drug resistant cases (MDR), treated with regimen A or B (for convenience, notation only includes 'a'), continue infectious:

$$\frac{dT^I_{Ma}}{dt}[1] = t_{CD}(1-p_{DST})p_{incM}I^{CD}_{M} - (t_{ph1}+\mu+\mu_T+\kappa)\ T^I_{Ma}[1] + t_{ph5}(1-p_{DST})p_{incM}P_M$$

$$\frac{dT^I_{Ma}}{dt}[2] = t_{ph1}r_{MDR}T^I_{Ra}[1] + t_{DST}rT^I_{Rb}[1] + t_{ph1}T^I_{Ma}[1] - (t_{ph2}+\mu+\mu_T+\kappa)\ T^I_{Ma}[2]$$

$$\frac{dT^I_{Ma}}{dt}[3] = t_{ph2}\left[r_{MDR}T^I_{Ra}[2] + T^I_{Ma}[2]\right] - (t_{ph3}+\mu+\mu_T+\kappa)\ T^I_{Ma}[3]$$

$$\frac{dT^I_{Ma}}{dt}[4] = t_{ph3}\left[r_{MDR}T^I_{Ra}[3] + T^I_{Ma}[3]\right] - (t_{ph4}+\mu+\mu_T+\kappa)\ T^I_{Ma}[4]$$

Multiple drug resistant cases (MDR), treated with regimen C, continue infectious and fail treatment:

$$\frac{dT^I_{Mc}}{dt}[1] = t_{CD}p_{DST}p_{incM}I^{CD}_{M} - (t_{DST}+\mu+\mu_T+\kappa)\ T^I_{Mc}[1] + t_{ph5}p_{DST}p_{incM}P_M$$

$$\frac{dT^{I}_{Mc}}{dt}[2] = t_{DST} T^{I}_{Mc}[1] - (t_{ph1} + \mu + \mu_T + \kappa)\ T^{I}_{Mc}[2]$$

$$\frac{dT^{I}_{Mc}}{dt}[3] = (1-\theta_{Mc})\ t_{ph1}\ T^{I}_{Mc}[2] - (t_{ph2} + \mu + \mu_T + \kappa)\ T^{I}_{Mc}[3]$$

$$\frac{dT^{I}_{Mc}}{dt}[4] = t_{ph2}\ T^{I}_{Mc}[3] - (t_{ph3} + \mu + \mu_T + \kappa)\ T^{I}_{Mc}[4]$$

$$\frac{dT^{I}_{Mc}}{dt}[5] = t_{ph3}\ T^{I}_{Mc}[4] - (t_{ph4} + \mu + \mu_T + \kappa)\ T^{I}_{Mc}[5]$$

Multiple drug resistant cases (MDR), treated with regimen C, become non-infectious through effective treatment:

$$\frac{dT^{NI}_{Mc}}{dt}[1] = 0$$

$$\frac{dT^{NI}_{Mc}}{dt}[2] = \theta_{Mc}\, t_{ph1}\ T^{I}_{Mc}[2] - (t_{ph2} + \mu + \mu_T + \kappa)\ T^{NI}_{Mc}[2]$$

$$\frac{dI^{NI}_{Mc}[3]}{dt} = t_{pl2}\ T^{NI}_{Mc}[2] - (t_{pl3}+\mu+\mu_T+\kappa)\ T^{NI}_{Mc}[3]$$

$$\frac{dI^{NI}_{Mc}[4]}{dt} = t_{pl3}\ T^{NI}_{Mc}[3] - (t_{pl4}+\mu+\mu_T+\kappa)\ T^{NI}_{Mc}[4]$$

MDR cases which are either cured (C) or have failed treatment (P, previously treated):

$$\frac{dC_M}{dt} = t_{pl4}\left[\ (1-w^{I}_{Mc})\ T^{I}_{Mc}[4] + (1-w^{NI}_{Mc})\ T^{NI}_{Mc}[4]\ \right] + \kappa(I^{CD}_{M} + T^{I}_{Ma}[1.4] + T^{I}_{Mc}[1.5] + T^{NI}_{Ma}[1.4] + I_M + U_M + P_M) - (\mu+\gamma p\lambda)C_M$$

$$\frac{dP_M}{dt} = t_{pl4}\left[T^{I}_{Ma}[4] + w^{I}_{Mc}\ T^{I}_{Mc}[4] + w^{NI}_{Mc}\ T^{NI}_{Mc}[4]\right] - (t_{pl5}+\mu+\mu_T+\kappa)P_M$$

$$L = L_S + L_R + L_M$$
$$I = I_S + I_R + I_M$$
$$T = T_S + T_R + T_M$$
$$N = X + L + I + T$$

Table 12-1. Parameter values for model presented in chapter 7. Rates are given in per month unless otherwise indicated.

Parameter	Explanation	Range		Source
		INTRINSIC PARAMETERS		
c	Contact rate, strain specific	1 – 1.5	About 1 effective contact / case-month	
p	Probability direct progression	0.10		
v	Reactivation rate	0.0015 / year		
u	Mortality rate	0.015 / year		
u_T	TB-mortality rate	0.05 / year		
k	Self-cure rate	0.5 /year		
r_1	AR rate	5-15%	Acquired resistance rate for infectious DS case under regimen A, or infectious DR case under regimen B	
r_2	AR rate	0-3%	Acquired resistance rate for non-infectious DS case under regimen A	
r_{MDR}	AR rate	20-60%	Acquired resistance rate for infectious DR case under regimen A	
γ	Reinfection susceptibility	~50%		
inf	Proportion developing infectious disease	40-60%		
		CONTROL PARAMETERS		
Φ_{Sa}	TcSa			
Φ_{Ra}	TcRa	Assume 0%		
Φ_{Rb}	TcRb			
Φ_{Ma}	TcMa	Assume 0%		
Φ_{Mc}	TcMc			
w_1	Relsa			
w_2	RelsaI			
w_3	Relrb			
w_4	RelrbI			
CDR	Case detection rate			
t_{CD}	Time to detect			
p_{DST}	Proportion new I_R			

	cases receiving DST			
t_{DST}	Time to DST	0 – 2 months		
t_1	For bookkeeping: time to remove from new incidence class	1 week		
$t_{ph1..4}$	Time each treatment phase	Phase 1: 2 weeks Phase 2: 3 months Phase 3: 3 months Phase 4: 3 months		
t_{ph5}	Time to re-treatment	1 year		
STATE VARIABLES				
Symbol	Explanation	By strain type	Infectious?	
X	Susceptible		-	
L	Latent	L_S , L_R, L_M	-	
NI	Non-infectious disease		-	
I	New infectious cases	I_S , I_R, I_M	+	
U	Untreated infectious cases	U_S ,U_R,U_M	+	
T	Treated cases		+ / -	
P	Previously failed treatment	P_S , P_R, P_M	+	
C	Cured	C_S , C_R, C_M	-	
TREATMENT STATE VARIABLES				
Symbol	Description	Strain type	Infectious	Treatment regimen
T_{SIa}	Sensitive infectious cases receiving treatment a	S	+	a
T_{Sa}	Sensitive non-infectious cases receiving treatment a	S	-	a
T_{RIa}	Resistant infectious cases receiving treatment a	R	+	a
T_{RIb}	Resistant infectious cases receiving treatment b	R	+	b
T_{Rb}	Resistant non-infectious cases receiving treatment b	R	-	b
T_{MIa}	MDR infectious cases receiving treatment a (or b)	M	+	a or b
T_{MIc}	MDR infectious cases receiving treatment c	M	+	c
T_{MIc}	MDR non-infectious cases receiving treatment c	M	-	c

13. Appendix 3

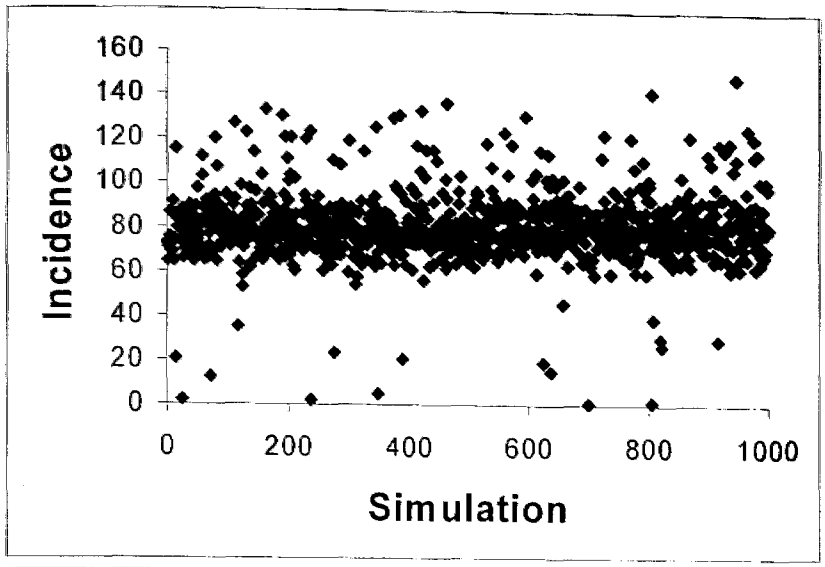
Sensitivity Analysis Scenarios for Chapter 6

Simulation	Treatment parameters	Intrinsic parameters	Reinfection	Incidence rate (Cases per 100,000)			Prevalence of resistance (% of cases resistant)		
				Average	Min	Max	Average	Min	Max
1	None	Varied	None	81	0.7	149	-----	-----	-----
2	None	Varied	Varied	1140	0.1	4403	-----	-----	-----
3	None	Varied	Constant	1540	0.8	3217	-----	-----	-----
4	Control parameters varied	Varied	Varied	355	31	2899	17%	0%	96%
5	Control parameters varied	Varied	Constant	364	63	1582	18%	0%	89%
6	Notification rate varied over wide range	Varied	Constant	320	23	1601	8%	0%	22%
7	Good control	Varied	Constant	157	12	299	3%	1%	5%
8	Poor control	Varied	Constant	379	44	791	37%	9%	67%

Table 13-1. **Summary of sensitivity analysis simulations.** ***Comparison of Monte Carlo simulations in sensitivity analysis.*** Each simulation consists of 1000 Monte Carlo runs of the model, with the intrinsic and control parameters varied between runs as given. The results of these simulations are shown as the final incidence of disease and prevalence of resistance.

SCENARIO 1: No treatment, no reinfection

A.



Model Outcome	Average	Min	Max
Incidence	81	1	149
Prevalence Resistance	0%	0%	0%
DOI-DS	82	1	150

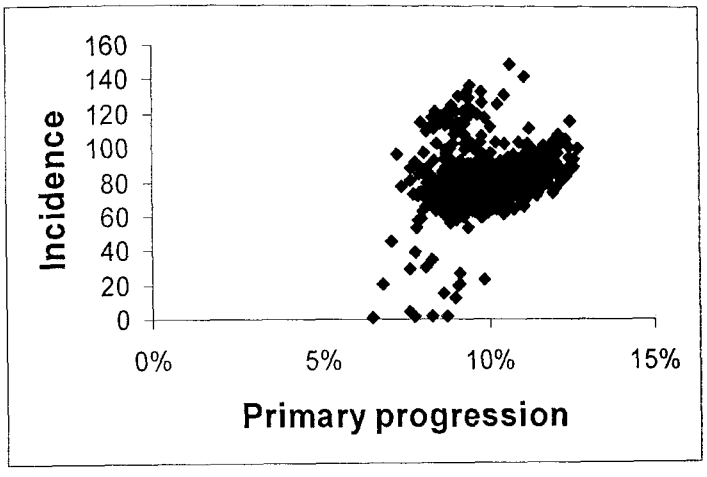
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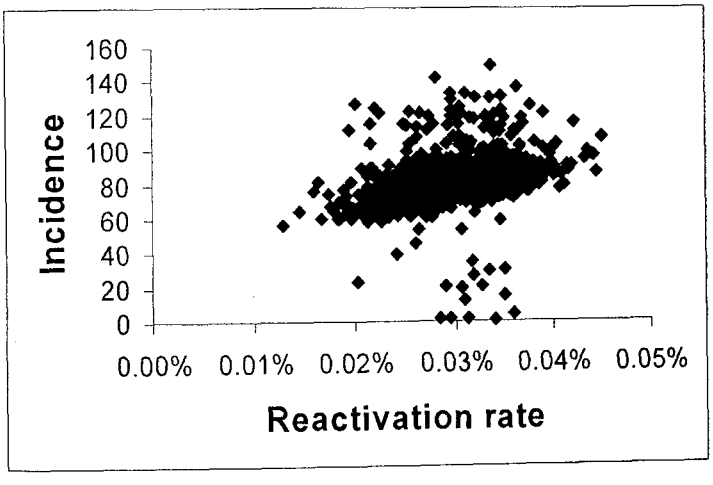
Parameter	Parameter explanation	Mean	Variance
Intrinsic parameters			
μ	Mortality rate	0.015	0.01
μ_T	TB disease specific mortality rate	0.2	0.01
k	Self-cure rate	0.1	0.005
ECR	Effective contact rate	10	1
i	Proportion progress to infectious disease	0.55	0.01
p	Proportion progress to primary disease	0.1	0.01
v	Endogenous reactivation rate	0.03%	0.001%
γ	Susceptibility to reinfection	0	0
f	Resistance transmission reduced by	0	0
w	Relapse rate	0.05	0.01
Treatment parameters			
nr	Notification rate	0	0
Φ_S	Treatment success rate for DS cases	0	0
Φ_R	Treatment success rate for DR cases	0	0
r	Acquired resistance rate	0	0

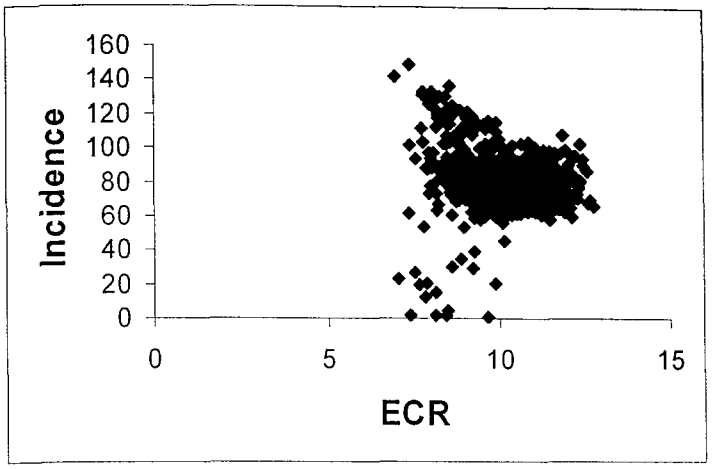
C.

Parameter	Parameter explanation	r^2 Prevalence disease
Intrinsic parameters		
μ	Mortality rate	**3.0%**
μ_T	TB disease specific mortality rate	0.1%
k	Self-cure rate	0.0%
ECR	Effective contact rate	**2.1%**
i	Proportion progress to infectious disease	0.0%
p	Proportion progress to primary disease	**7.1%**
v	Endogenous reactivation rate	**9.5%**
w	Relapse rate	0.0%
γ	Susceptibility to reinfection	0.0%

D.



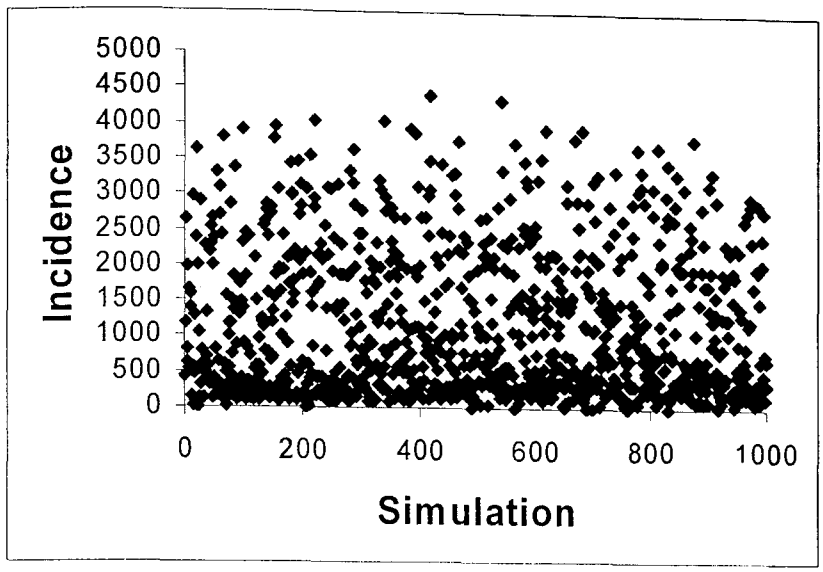




Scenario 1. *Model sensitivity under no drug treatment.* The results of 1000 Monte Carlo simulations run for ten years under no treatment (notification rate set to 0) are shown as the final incidence of infectious disease (A). Model parameters were randomly generated in each simulation, assumed to have a normal distribution with mean and variance as given in (B). The Pearson squared correlation (r^2) calculated in (C) reflects the proportion of variance in model outcome explained by variance in each parameter. Scatterplots for the input parameters which most influenced the model outcome of disease incidence (as new cases per 100,000) are shown in (D).

SCENARIO 2: No treatment, reinfection varied

A.



Model Outcome	Average	Min	Max
Incidence	1140	0	4403
Prevalence Resistance	0%	0%	0%
DOI-DS	2.21	1.95	2.46

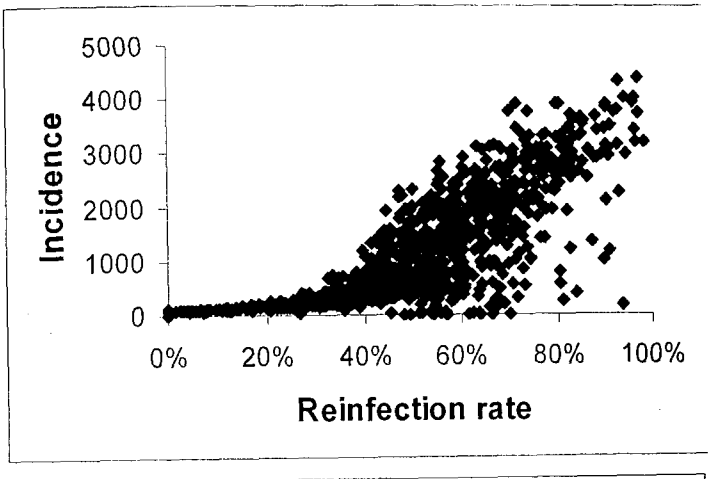
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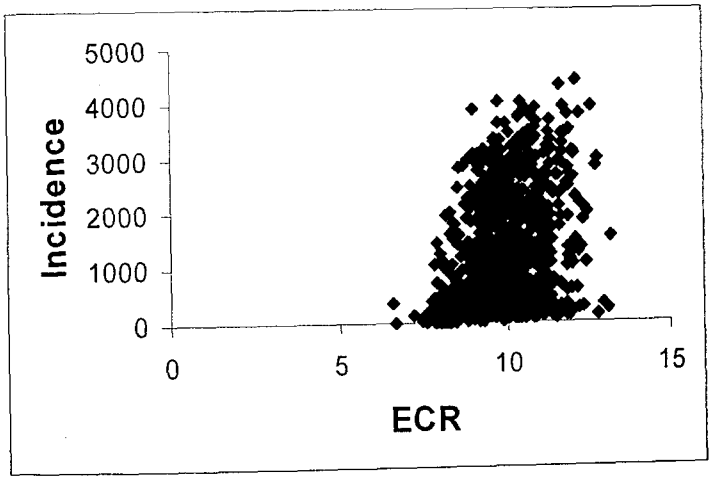
Parameter	Parameter explanation	Mean	Variance
Intrinsic parameters			
μ	Mortality rate	0.015	0.01
μ_T	TB disease specific mortality rate	0.2	0.01
k	Self-cure rate	0.1	0.005
ECR	Effective contact rate	10	1
i	Proportion progress to infectious disease	0.55	0.01
p	Proportion progress to primary disease	0.1	0.01
v	Endogenous reactivation rate	0.03%	0.001%
γ	Susceptibility to reinfection	0.5	0.2
f	Resistance transmission reduced by	0	0
w	Relapse rate	0.05	0.01
Treatment parameters			
nr	Notification rate	0	0
Φ_S	Treatment success rate for DS cases	0	0
Φ_R	Treatment success rate for DR cases	0	0
r	Acquired resistance rate	0	0

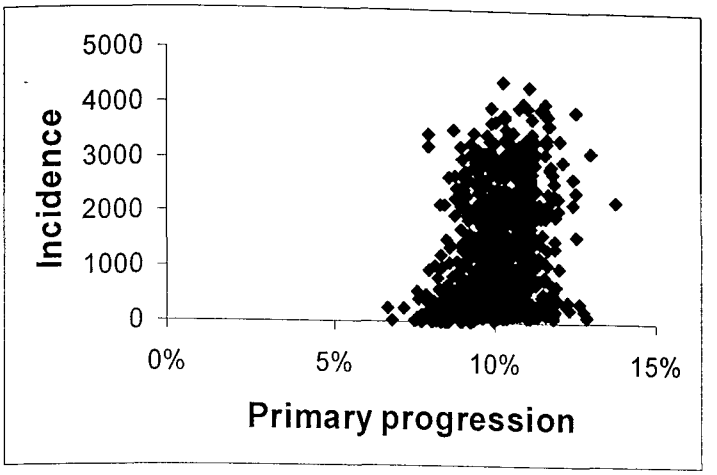
C.

Parameter	Parameter explanation	r^2 Incidence
Intrinsic parameters		
μ	Mortality rate	0.0%
μ_T	TB disease specific mortality rate	**2.2%**
k	Self-cure rate	0.0%
ECR	Effective contact rate	**8.8%**
i	Proportion progress to infectious disease	0.3%
p	Proportion progress to primary disease	**11.0%**
v	Endogenous reactivation rate	0.0%
w	Relapse rate	1.3%
γ	Susceptibility to reinfection	**62.7%**

D.



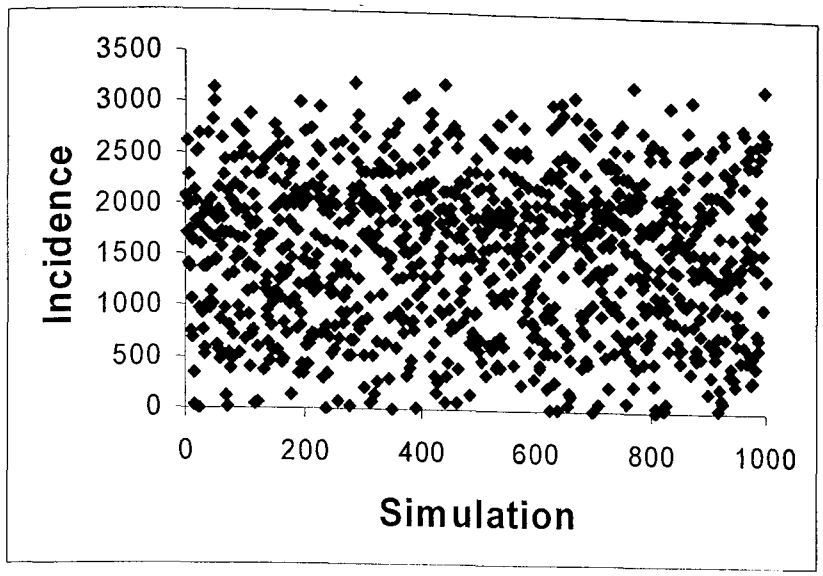




Scenario 2. *Model sensitivity under no drug treatment, and widely varied reinfection rate.* The results of 1000 Monte Carlo simulations run for ten years under no treatment (notification rate set to 0) are shown as the final incidence of infectious disease (A). Model parameters were randomly generated in each simulation, assumed to have a normal distribution with mean and variance as given in (B). The Pearson squared correlation (r^2) calculated in (C) reflects the proportion of variance in model outcome explained by variance in each parameter. Scatterplots for the input parameters which most influenced the model outcome of disease incidence are shown in (D).

SCENARIO 3: No treatment, reinfection constant

A.



Model Outcome	Average	Min	Max
Incidence	1540	1	3217
Prevalence Resistance	0%	0%	0%
DOI-DS	2.21	1.95	2.50

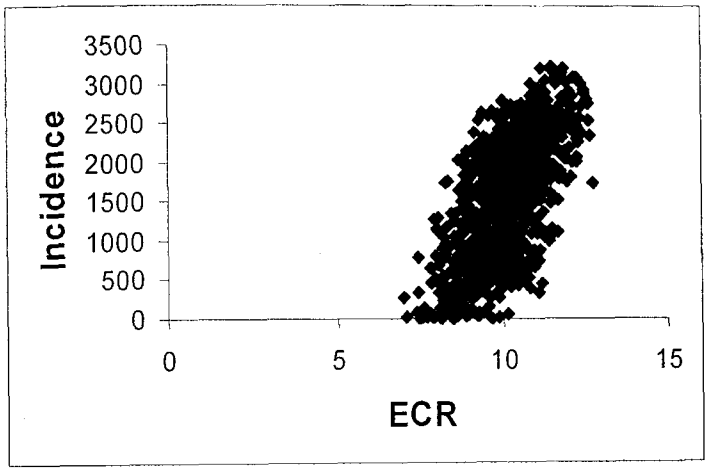
B.

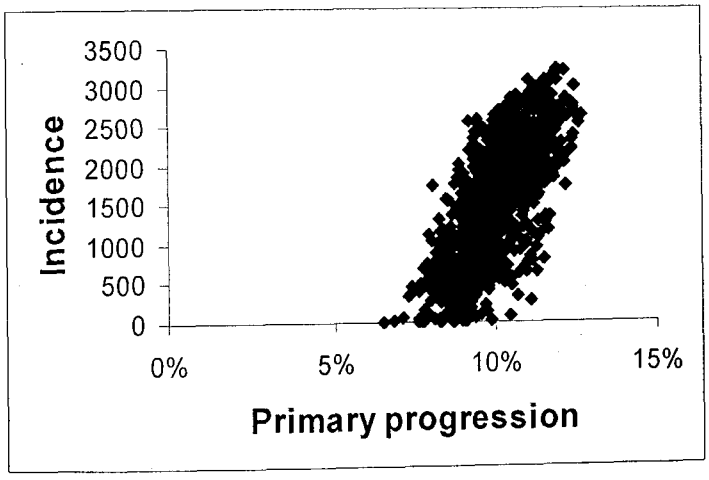
Parameter	Parameter explanation	Mean	Variance
Intrinsic parameters			
μ	Mortality rate	0.015	0.01
μ_T	TB disease specific mortality rate	0.2	0.01
k	Self-cure rate	0.1	0.005
ECR	Effective contact rate	10	1
i	Proportion progress to infectious disease	0.55	0.01
p	Proportion progress to primary disease	0.1	0.01
v	Endogenous reactivation rate	0.03%	0.001%
γ	Susceptibility to reinfection	0.60	0.01
f	Resistance transmission reduced by	0	0
w	Relapse rate	0.05	0.01
Treatment parameters			
nr	Notification rate	0	0
Φ_S	Treatment success rate for DS cases	0	0
Φ_R	Treatment success rate for DR cases	0	0
r	Acquired resistance rate	0	0

C.

Parameter	Parameter explanation	r^2 Incidence
Intrinsic parameters		
μ	Mortality rate	0.0%
μ_T	TB disease specific mortality rate	**4.5%**
k	Self-cure rate	0.0%
ECR	Effective contact rate	**40.4%**
i	Proportion progress to infectious disease	0.5%
p	Proportion progress to primary disease	**48.4%**
v	Endogenous reactivation rate	0.3%
w	Relapse rate	1.7%
γ	Susceptibility to reinfection	1.6%

D.

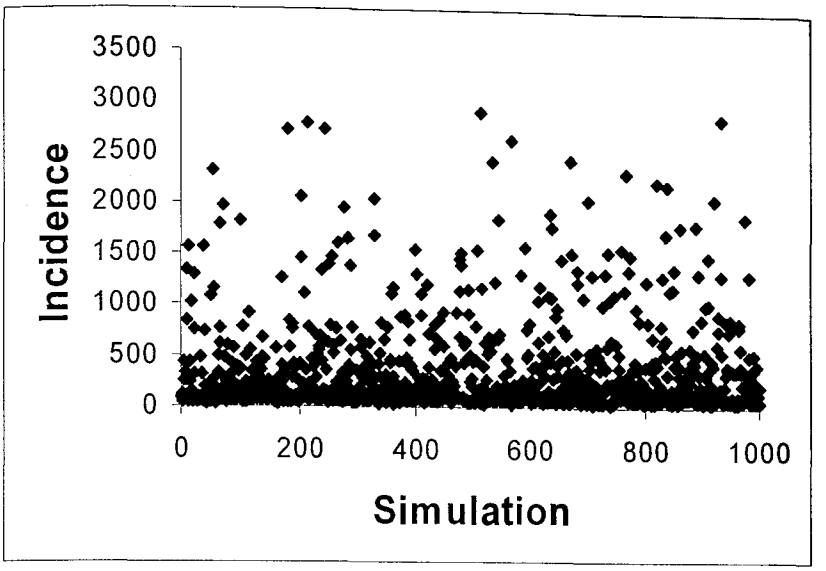


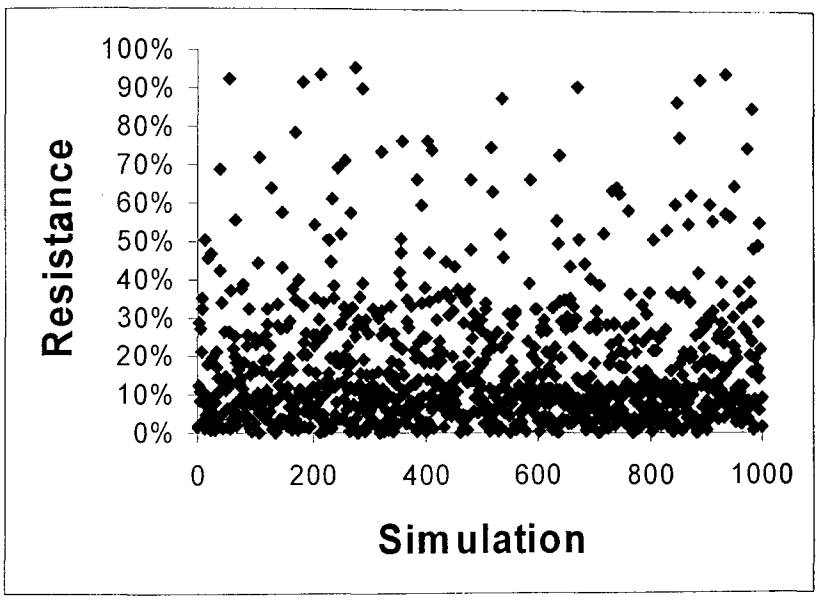


Scenario 3. *Model sensitivity under no drug treatment, and a constant reinfection rate.* The results of 1000 Monte Carlo simulations run for ten years under no treatment (notification rate set to 0) are shown as the final incidence of infectious disease (A). Model parameters were randomly generated in each simulation, assumed to have a normal distribution with mean and variance as given in (B). The Pearson squared correlation (r^2) calculated in (C) reflects the proportion of variance in model outcome explained by variance in each parameter. Scatterplots for the input parameters which most influenced the model outcome of disease incidence are shown in (D).

SCENARIO 4: Varied control, varied reinfection

A.





Model Outcome	Average	Min	Max
Incidence Rate	355	31	2899
Prevalence Resistance	17%	0%	96%
Incidence Rate- Drug sensitive	335	29	4106
Incidence rate- Drug resitant	135	0	5594
DOI-DS	1.03	0.55	2.22

DOI-DR	1.61	0.91	2.25

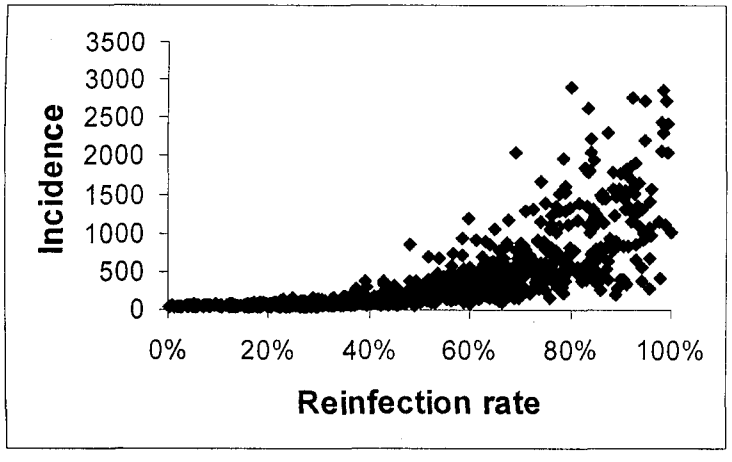
B.

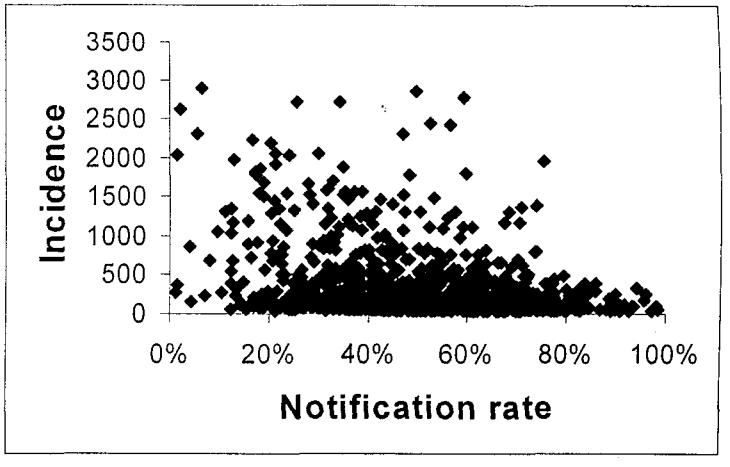
Parameter	Parameter explanation	Mean	Variance
Intrinsic parameters			
μ	Mortality rate	0.015	0.001
μ_T	TB disease specific mortality rate	0.2	0.01
k	Self-cure rate	0.1	0.1
ECR	Effective contact rate	10	0.5
i	Proportion progress to infectious disease	0.55	0.01
p	Proportion progress to primary disease	0.1	0.005
v	Endogenous reactivation rate	0.0003	0.00005
γ	Susceptibility to reinfection	0.5	0.3
f	Resistance transmission reduced by	0.1	0.1
w	Relapse rate	0.03	0.005
Treatment parameters			
nr	Notification rate	0.5	0.2
Φ_S	Treatment success rate for DS cases	0.9	0.2
Φ_R	Treatment success rate for DR cases	0.25	0.1
r	Acquired resistance rate	0.08	0.03

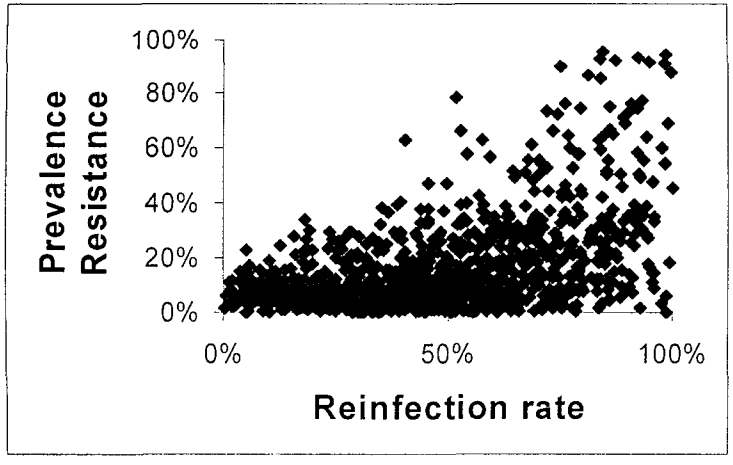
C.

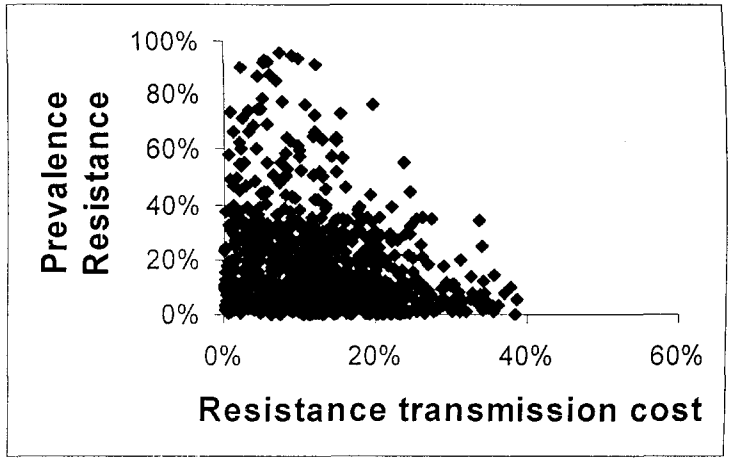
Parameter	Parameter explanation	r^2 Incidence	r^2 Prevalence of Resistance
Intrinsic parameters			
μ	Mortality rate	0.2%	0.4%
μ_T	TB disease specific mortality rate	0.3%	0.3%
k	Self-cure rate	0.3%	0.1%
ECR	Effective contact rate	0.0%	0.3%
i	Proportion progress to infectious disease	0.2%	0.3%
p	Proportion progress to primary disease	1.6%	0.7%
v	Endogenous reactivation rate	0.5%	0.2%
γ	Susceptibility to reinfection	50.8%	23.5%
f	Resistance transmission reduced by	1.0%	6.2%
w	Relapse rate	1.0%	0.0%
Treatment parameters			
nr	Notification rate	9.3%	4.7%
Φ_S	Treatment success rate for DS cases	1.3%	12.7%
Φ_R	Treatment success rate for DR cases	0.8%	13.3%
r	Acquired resistance rate	0.1%	9.1%

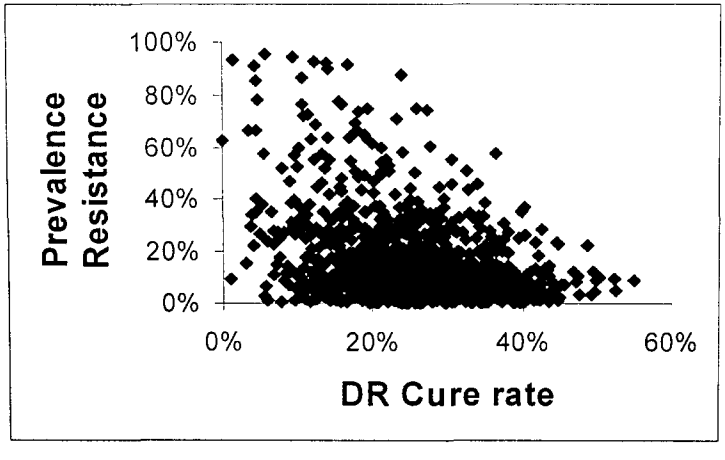
D.

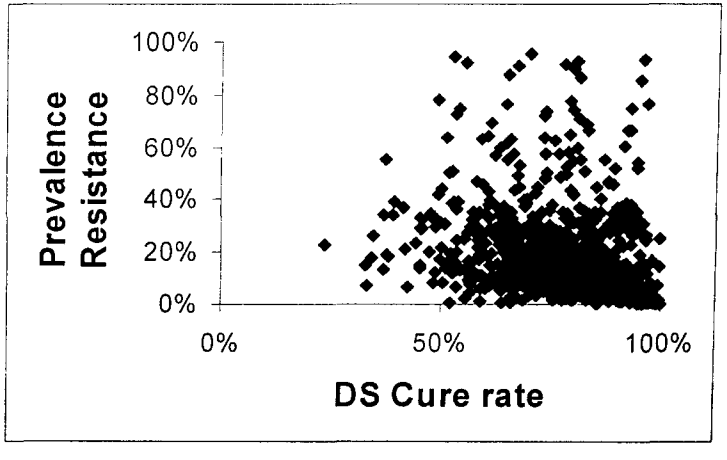

Incidence
3500
3000
2500
2000
1500
1000
500
0
0%
20%
40%
60%
80%
100%
Reinfection rate

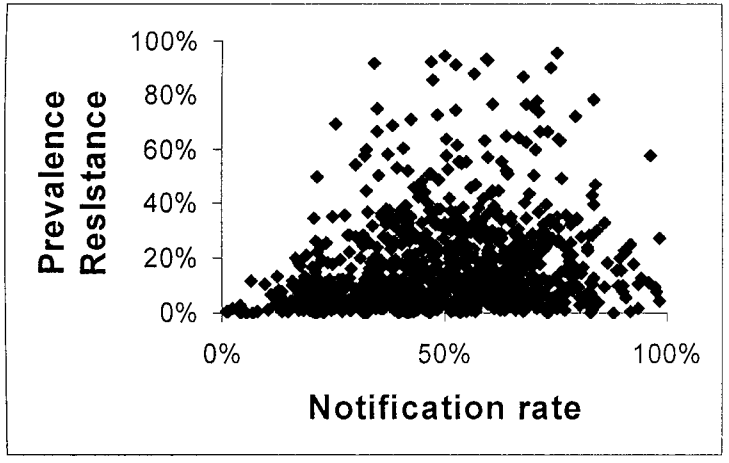

Incidence
3500
3000
2500
2000
1500
1000
500
0
0%
20%
40%
60%
80%
100%
Notification rate

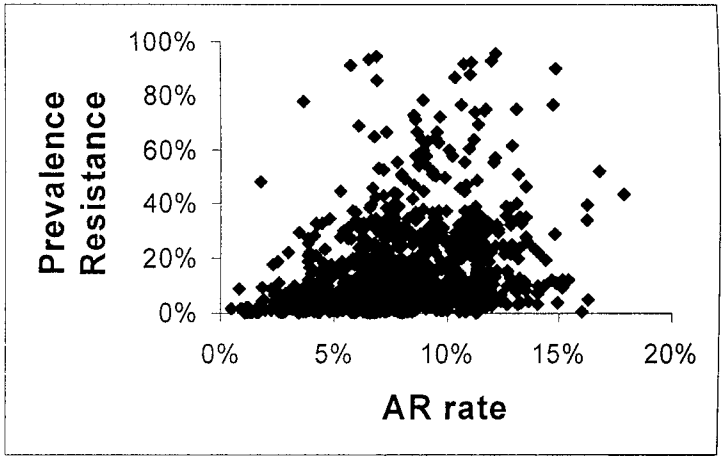

Prevalence Resistance
100%
80%
60%
40%
20%
0%
0%
50%
100%
Reinfection rate


Prevalence Resistance
100%
80%
60%
40%
20%
0%
0%
20%
40%
60%
Resistance transmission cost


Prevalence Resistance
100%
80%
60%
40%
20%
0%
0%
20%
40%
60%
DR Cure rate


Prevalence Resistance
100%
80%
60%
40%
20%
0%
0%
50%
100%
DS Cure rate

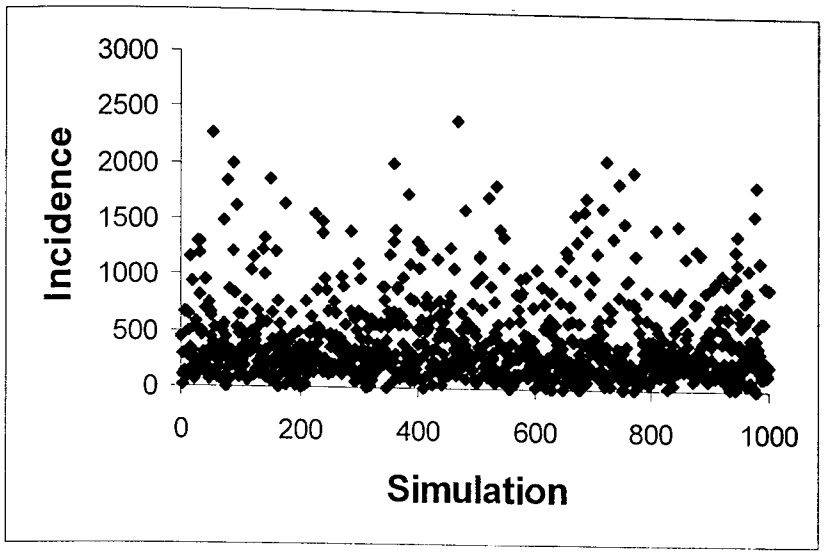


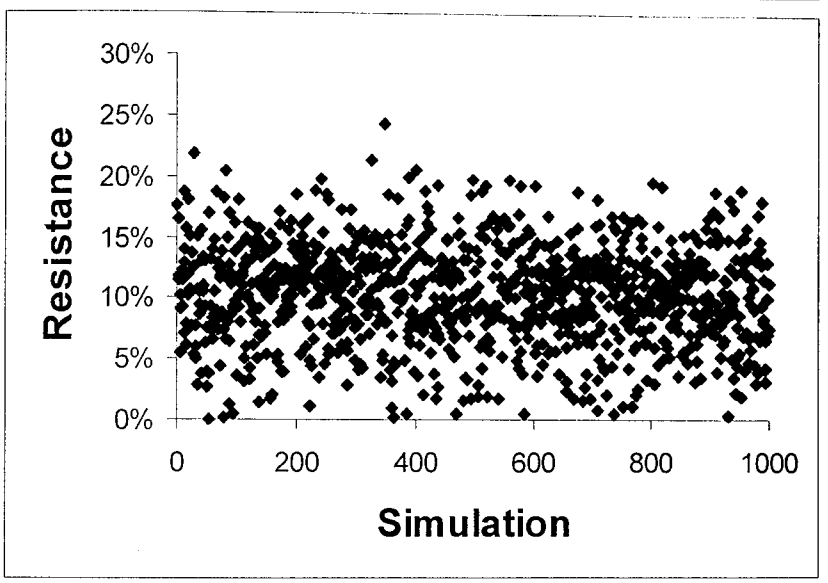


Scenario 4. *Model sensitivity under widely varied control and reinfenction rates.* The results of 1000 Monte Carlo simulations run for ten years under no treatment (notification rate set to 0) are shown as the final incidence of infectious disease (A). Model parameters were randomly generated in each simulation, assumed to have a normal distribution with mean and variance as given in (B). The Pearson squared correlation (r^2) calculated in (C) reflects the proportion of variance in model outcome explained by variance in each parameter. Scatterplots for the model outcome of incidence are shown in (D) and resistance prevalence in (E).

SCENARIO 5: Varied control

A.





Model Outcome	Average	Min	Max
Incidence Rate	364	63	1582
Prevalence Resistance	18%	0%	89%
Incidence Rate- Drug sensitive	357	59	2241
Incidence rate- Drug resitant	75	0	1189
DOI-DS	1.04	0.55	2.22
DOI-DR	1.62	0.92	2.33

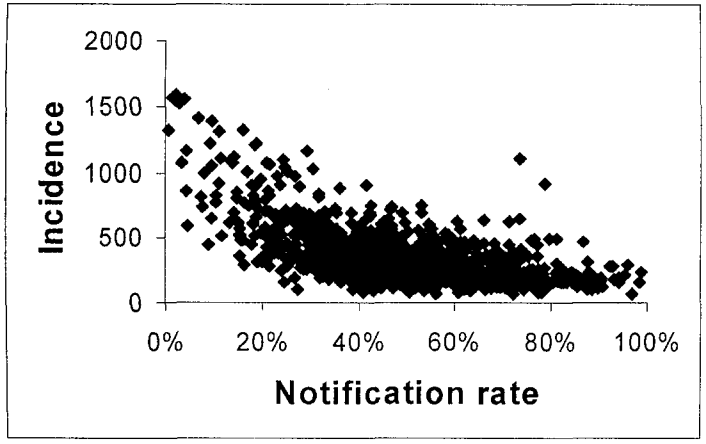
B.

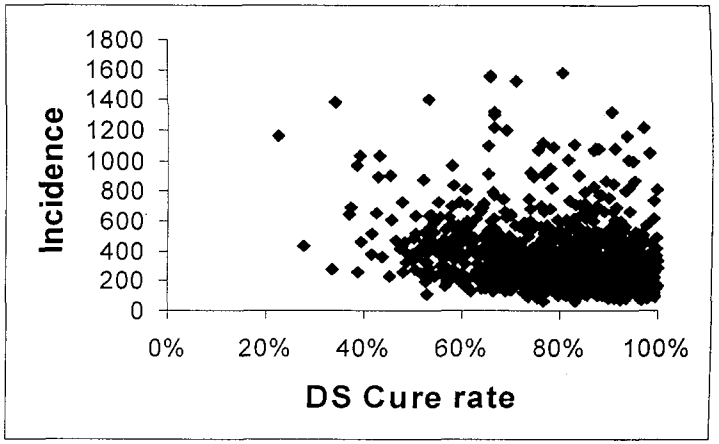
Parameter	Parameter explanation	Mean	Variance
Intrinsic parameters			
μ	Mortality rate	0.015	0.001
μ_T	TB disease specific mortality rate	0.2	0.01
k	Self-cure rate	0.1	0.1
ECR	Effective contact rate	10	0.5
i	Proportion progress to infectious disease	0.55	0.01
p	Proportion progress to primary disease	0.1	0.005
v	Endogenous reactivation rate	0.0003	0.00005
γ	Susceptibility to reinfection	0.6	0.01
f	Resistance transmission reduced by	0.1	0.1
w	Relapse rate	0.03	0.005
Treatment parameters			
nr	Notification rate	0.5	0.2
Φ_S	Treatment success rate for DS cases	0.9	0.2
Φ_R	Treatment success rate for DR cases	0.25	0.1
r	Acquired resistance rate	0.08	0.03

C.

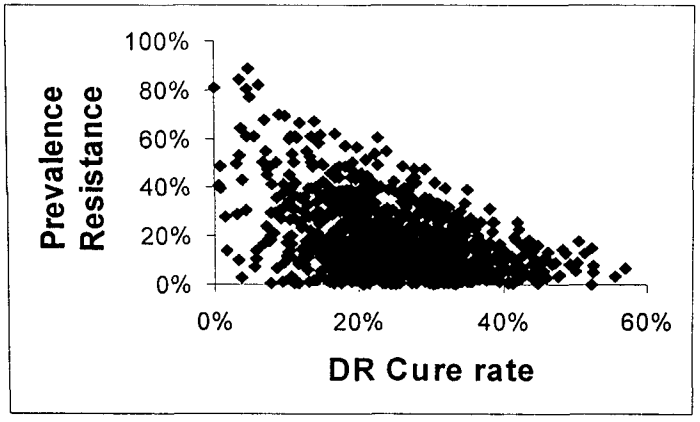
Parameter	Parameter explanation	r^2 Incidence	r^2 Prevalence of Resistance
Intrinsic parameters			
μ	Mortality rate	0.6%	0.0%
μ_T	TB disease specific mortality rate	2.4%	0.4%
k	Self-cure rate	0.2%	0.2%
ECR	Effective contact rate	8.2%	0.3%
i	Proportion progress to infectious disease	0.0%	0.0%
p	Proportion progress to primary disease	10.0%	1.2%
v	Endogenous reactivation rate	0.6%	0.1%
γ	Susceptibility to reinfection	0.6%	0.1%
f	Resistance transmission reduced by	0.9%	9.6%
w	Relapse rate	4.4%	0.3%
Treatment parameters			
nr	Notification rate	37.6%	9.8%
Φ_S	Treatment success rate for DS cases	7.2%	27.3%
Φ_R	Treatment success rate for DR cases	0.8%	20.4%
r	Acquired resistance rate	0.0%	14.2%

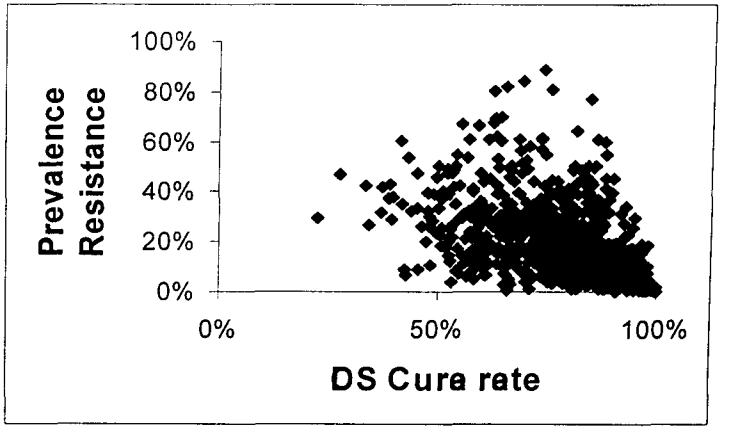
D.

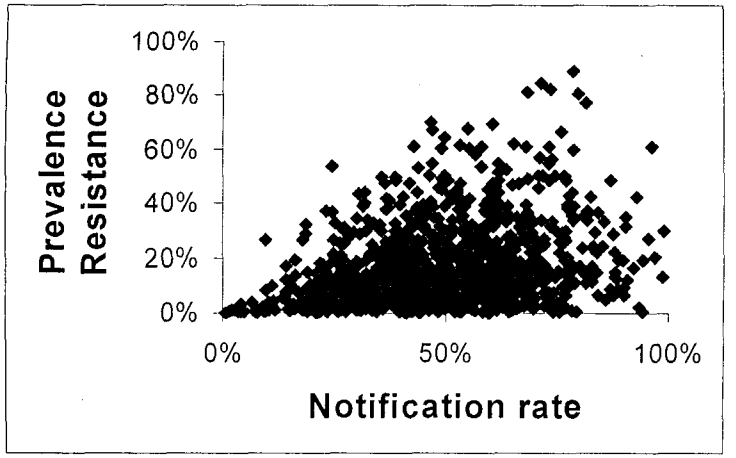

Incidence
2000
1500
1000
500
0
0%
20%
40%
60%
80%
100%
Notification rate

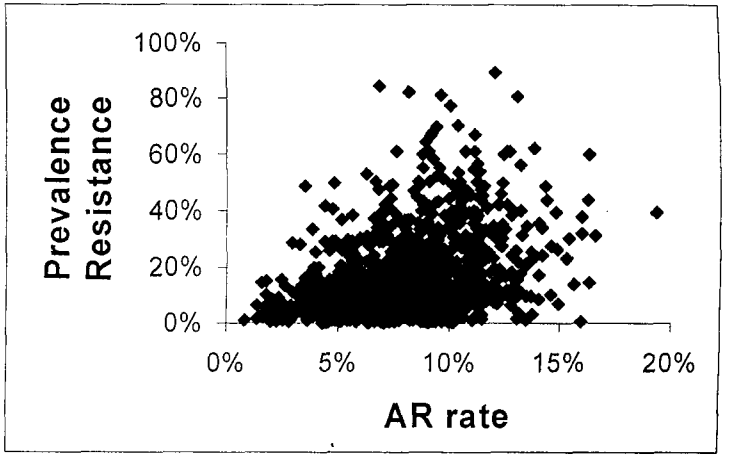

Incidence
1800
1600
1400
1200
1000
800
600
400
200
0
0%
20%
40%
60%
80%
100%
DS Cure rate

E.


Prevalence Resistance
100%
80%
60%
40%
20%
0%
0%
20%
40%
60%
DR Cure rate


Prevalence Resistance
100%
80%
60%
40%
20%
0%
0%
50%
100%
DS Cura rate

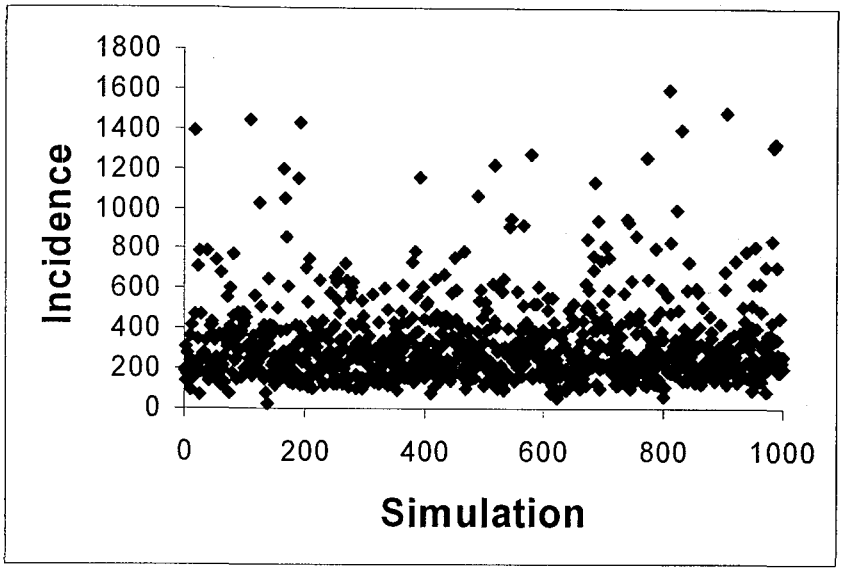


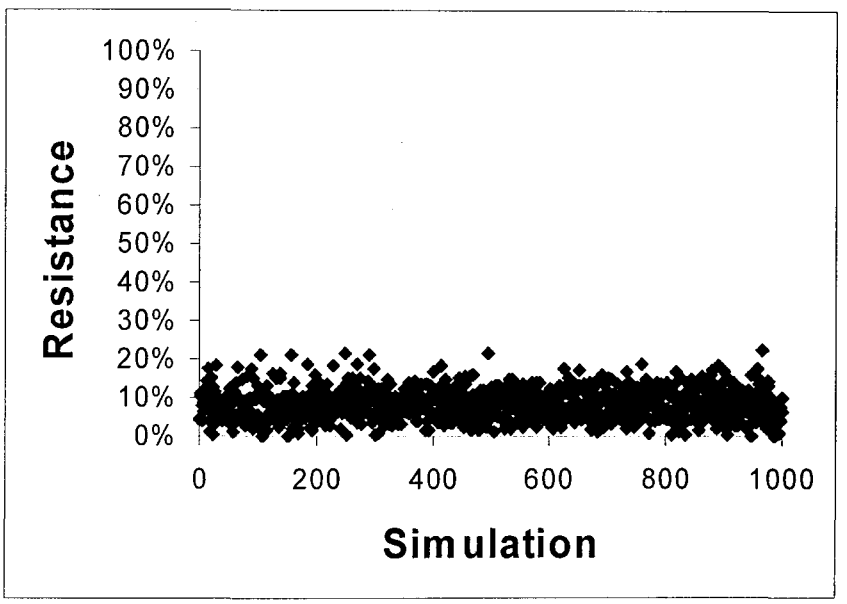


Scenario 5. *Model sensitivity under widely varied control, with a constant reinfenction rate.* The results of 1000 Monte Carlo simulations run for ten years under no treatment (notification rate set to 0) are shown as the final incidence of infectious disease (A). Model parameters were randomly generated in each simulation, assumed to have a normal distribution with mean and variance as given in (B). The Pearson squared correlation (r^2) calculated in (C) reflects the proportion of variance in model outcome explained by variance in each parameter. Scatterplots for the input parameters which most influenced the model outcome of incidence are shown in (D) and resistance prevalence in (E).

SCENARIO 6: Varied treatment rate

A.





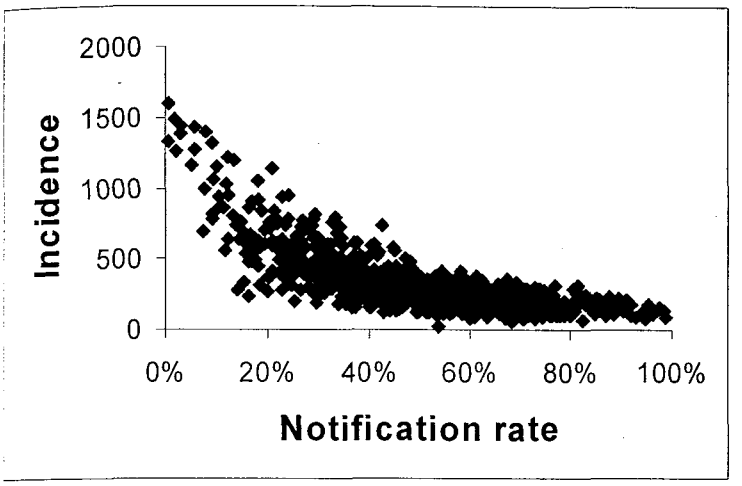
Model Outcome	Average	Min	Max
Incidence Rate	320	23	1601
Prevalence Resistance	8%	0%	22%
Incidence Rate- Drug sensitive	352	23	2027
Incidence rate- Drug resitant	26	0	166
DOI-DS	1.02	0.62	2.16
DOI-DR	1.27	0.85	2.22

B.

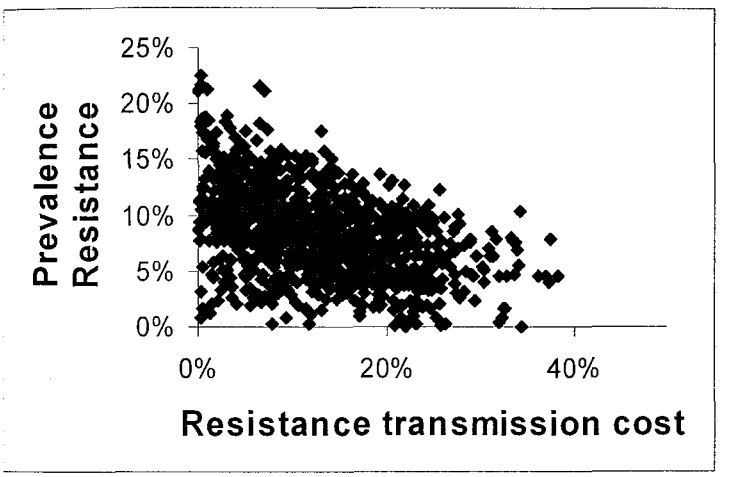
Parameter	Parameter explanation	Mean	Variance
Intrinsic parameters			
μ	Mortality rate	0.015	0.001
μ_T	TB disease specific mortality rate	0.2	0.01
k	Self-cure rate	0.1	0.1
ECR	Effective contact rate	10	0.5
i	Proportion progress to infectious disease	0.55	0.01
p	Proportion progress to primary disease	0.1	0.005
v	Endogenous reactivation rate	0.0003	0.00005
γ	Susceptibility to reinfection	0.6	0.01
f	Resistance transmission reduced by	0.1	0.1
w	Relapse rate	0.03	0.005
Treatment parameters			
nr	Notification rate	0.5	0.2
Φ_S	Treatment success rate for DS cases	0.8	0
Φ_R	Treatment success rate for DR cases	0.5	0
r	Acquired resistance rate	0.08	0.03

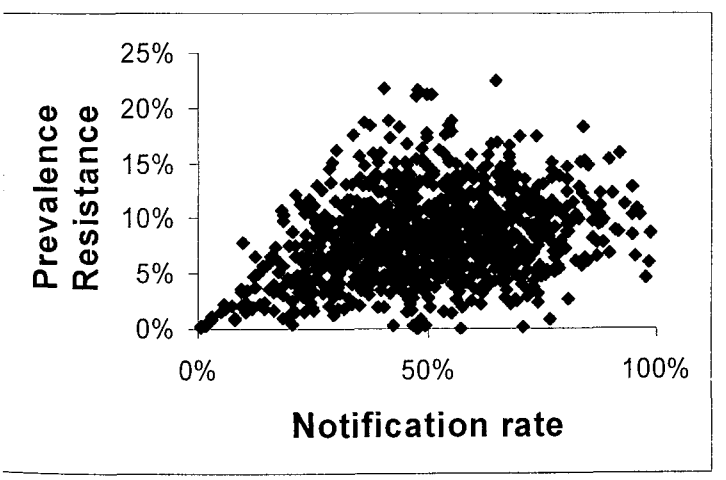
C.

Parameter	Parameter explanation	r^2 Incidence	r^2 Prevalence of Resistance
Intrinsic parameters			
μ	Mortality rate	0.1%	0.0%
μ_T	TB disease specific mortality rate	3.7%	0.5%
k	Self-cure rate	0.2%	0.2%
ECR	Effective contact rate	8.2%	0.5%
i	Proportion progress to infectious disease	0.3%	0.1%
p	Proportion progress to primary disease	6.1%	1.9%
v	Endogenous reactivation rate	0.1%	0.1%
γ	Susceptibility to reinfection	0.7%	0.3%
f	Resistance transmission reduced by	0.0%	20.4%
w	Relapse rate	4.6%	1.0%
Treatment parameters			
nr	Notification rate	52.4%	8.5%
Φ_S	Treatment success rate for DS cases	-----	-----
Φ_R	Treatment success rate for DR cases	-----	-----
r	Acquired resistance rate	0.0%	57.8%

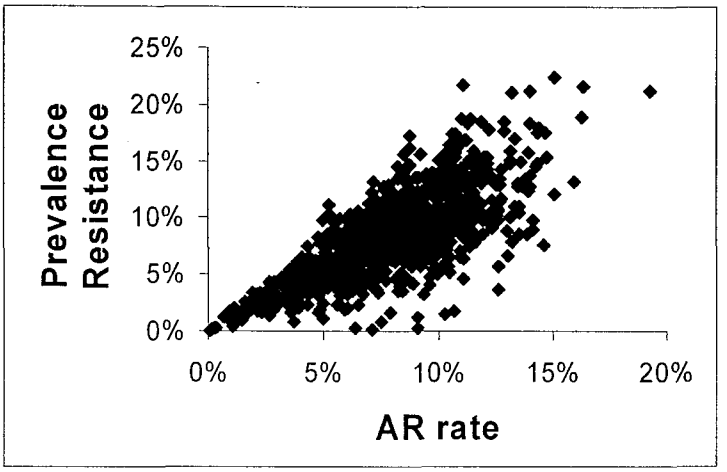

Incidence
2000
1500
1000
500
0
0%
20%
40%
60%
80%
100%
Notification rate

D.


Prevalence Resistance
25%
20%
15%
10%
5%
0%
0%
20%
40%
Resistance transmission cost


Prevalence Resistance
25%
20%
15%
10%
5%
0%
0%
50%
100%
Notification rate

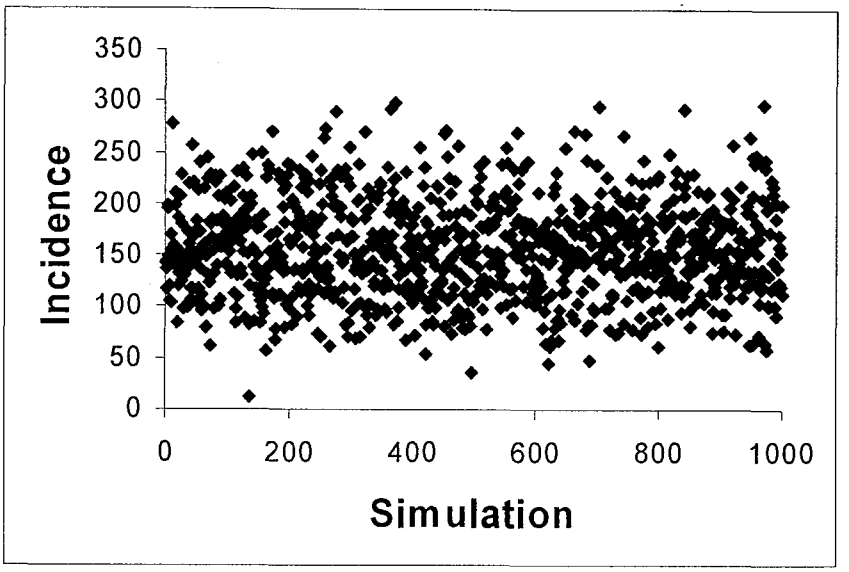
E.

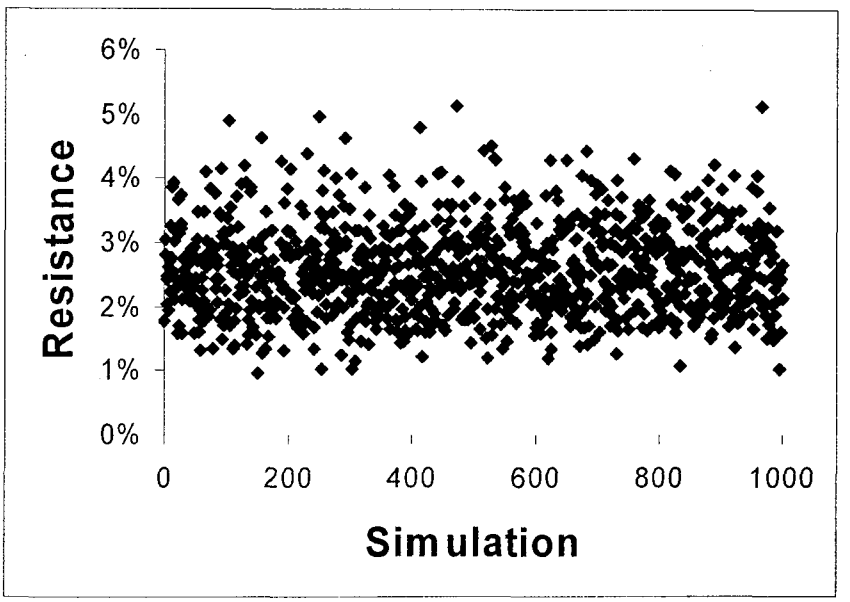


Scenario 6. *Model sensitivity under a varied treatment rates.* The results of 1000 Monte Carlo simulations run for ten years under no treatment (notification rate set to 0) are shown as the final incidence of infectious disease (A). Model parameters were randomly generated in each simulation, assumed to have a normal distribution with mean and variance as given in (B). The Pearson squared correlation (r^2) calculated in (C) reflects the proportion of variance in model outcome explained by variance in each parameter. Scatterplots for the input parameters which most influenced the model outcome of incidence are shown in (D) and resistance prevalence in (E).

SCENARIO 7: Good control

A.





Model Outcome	Average	Min	Max
Incidence Rate	157	12	299
Prevalence Resistance	3%	1%	5%
Incidence Rate- Drug sensitive	175	12	400
Incidence rate- Drug resitant	4	0	14
DOI-DS	0.70	0.68	0.73
DOI-DR	0.91	0.87	0.95

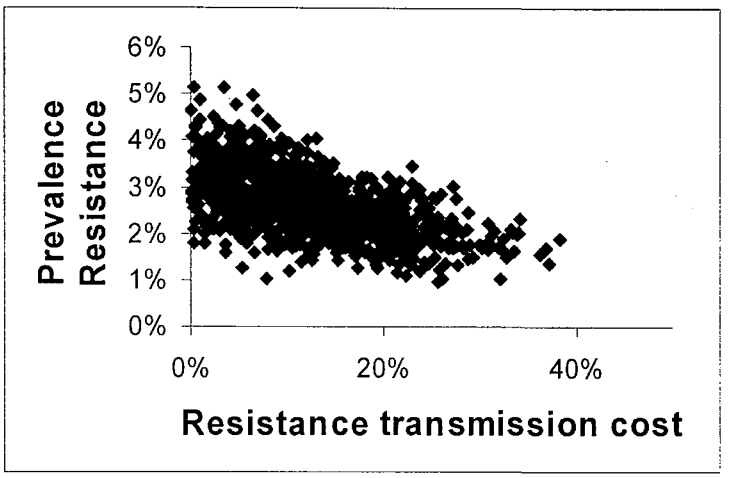
B.

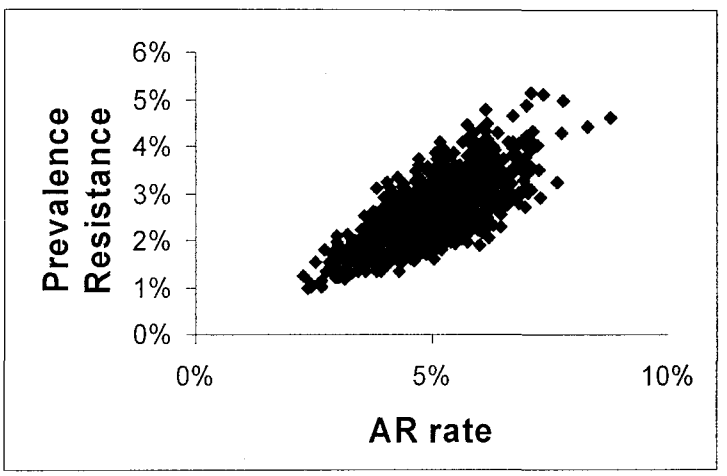
Parameter	Parameter explanation	Mean	Variance
Intrinsic parameters			
μ	Mortality rate	0.015	0.001
μ_T	TB disease specific mortality rate	0.2	0.01
k	Self-cure rate	0.1	0.1
ECR	Effective contact rate	10	0.5
i	Proportion progress to infectious disease	0.55	0.01
p	Proportion progress to primary disease	0.1	0.005
v	Endogenous reactivation rate	0.0003	0.00005
γ	Susceptibility to reinfection	0.6	0.01
f	Resistance transmission reduced by	0.1	0.1
w	Relapse rate	0.03	0.005
Treatment parameters			
nr	Notification rate	0.75	0
Φ_S	Treatment success rate for DS cases	0.9	0
Φ_R	Treatment success rate for DR cases	0.6	0
r	Acquired resistance rate	0.05	0.01

C.

Parameter	Parameter explanation	r^2 Incidence	r^2 Prevalence of Resistance
Intrinsic parameters			
μ	Mortality rate	0.6%	0.2%
μ_T	TB disease specific mortality rate	8.4%	2.0%
k	Self-cure rate	0.2%	0.8%
ECR	Effective contact rate	25.1%	0.9%
i	Proportion progress to infectious disease	1.5%	0.3%
p	Proportion progress to primary disease	34.0%	2.7%
v	Endogenous reactivation rate	1.0%	0.0%
γ	Susceptibility to reinfection	1.1%	0.5%
f	Resistance transmission reduced by	0.1%	31.4%
w	Relapse rate	19.4%	7.5%
Treatment parameters			
nr	Notification rate	-----	-----
Φ_S	Treatment success rate for DS cases	-----	-----
Φ_R	Treatment success rate for DR cases	-----	-----
r	Acquired resistance rate	0.0%	54.9%

D.



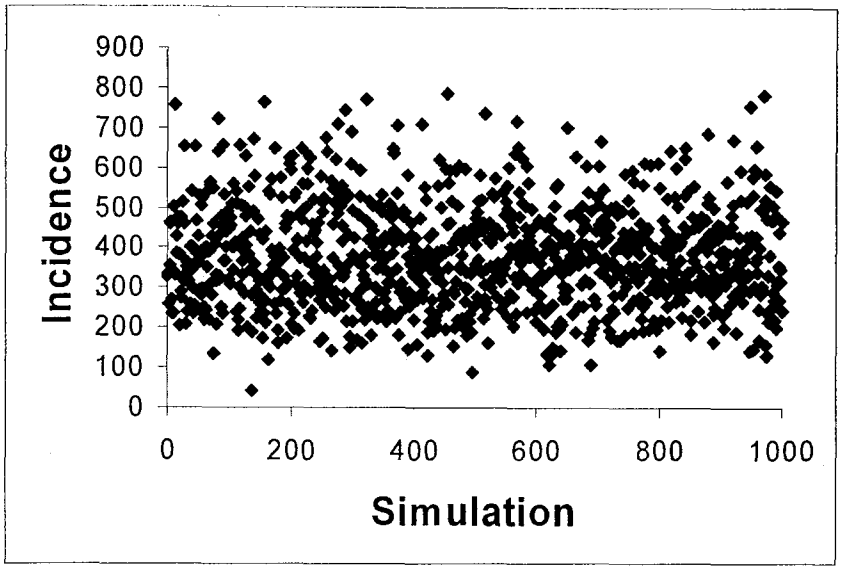


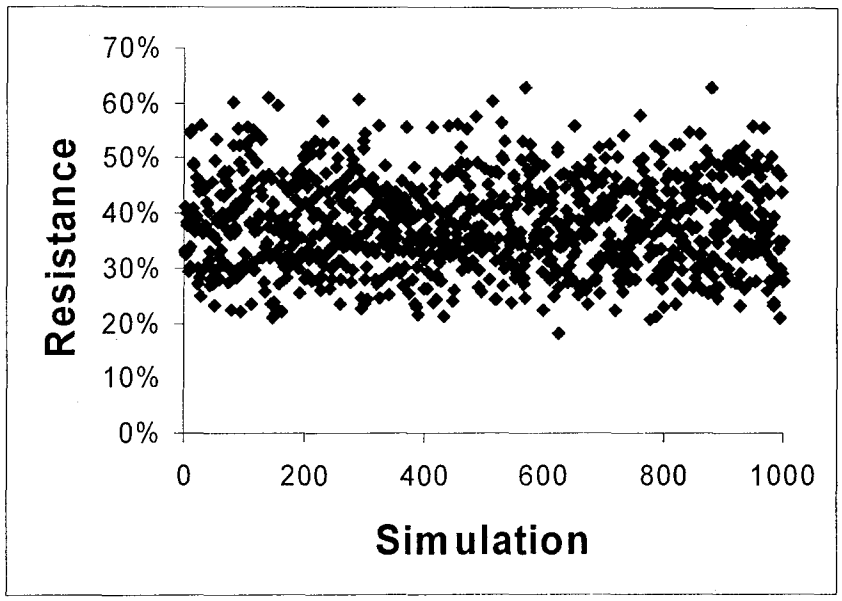
Scenario 7. *Model sensitivity under a good control program.* The results of 1000 Monte Carlo simulations run for ten years under no treatment (notification rate set to 0) are shown as the final incidence of infectious disease (A). Model parameters were randomly generated in each simulation, assumed to have a normal distribution with mean and variance as given in (B). The Pearson squared correlation (r^2) calculated in (C) reflects the proportion of variance in model outcome explained by variance in each parameter. Scatterplots for the input parameters which most influenced the model outcome of resistance prevalence are shown in (D).

A.

SCENARIO 8: Poor control

A.





Model Outcome	Average	Min	Max
Incidence Rate	379	44	791
Prevalence Resistance	37%	9%	67%
Incidence Rate- Drug sensitive	287	33	780
Incidence rate- Drug resitant	159	11	754
DOI-DS	1.13	1.07	1.20

DOI-DR	1.49	1.40	1.63

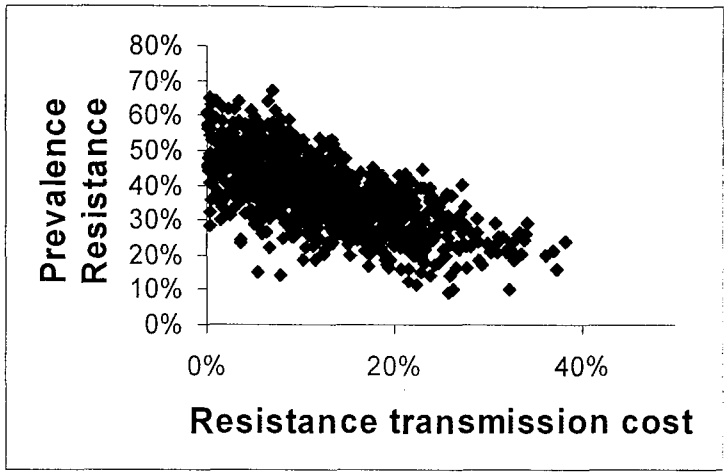
B.

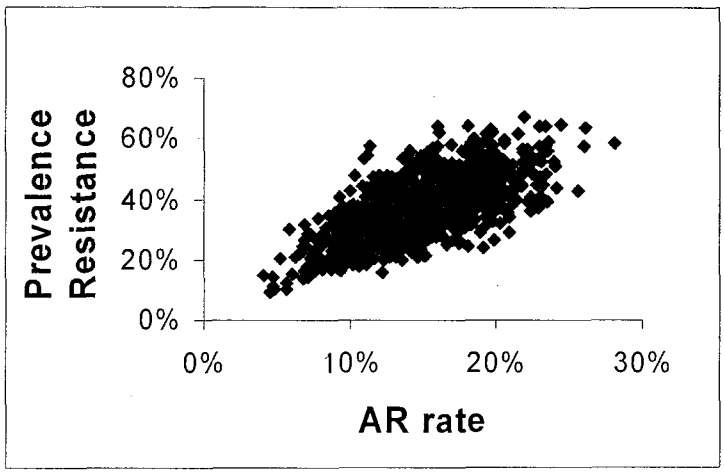
Parameter	Parameter explanation	Mean	Variance
Intrinsic parameters			
μ	Mortality rate	0.015	0.001
μ_T	TB disease specific mortality rate	0.2	0.01
k	Self-cure rate	0.1	0.1
ECR	Effective contact rate	10	0.5
i	Proportion progress to infectious disease	0.55	0.01
p	Proportion progress to primary disease	0.1	0.005
v	Endogenous reactivation rate	0.0003	0.00005
γ	Susceptibility to reinfection	0.6	0.01
f	Resistance transmission reduced by	0.1	0.1
w	Relapse rate	0.03	0.005
Treatment parameters			
nr	Notification rate	0.5	0
Φ_S	Treatment success rate for DS cases	0.6	0
Φ_R	Treatment success rate for DR cases	0.3	0
r	Acquired resistance rate	0.15	0.04

C.

Parameter	Parameter explanation	r^2 Incidence	r^2 Prevalence of Resistance
Intrinsic parameters			
μ	Mortality rate	0.3%	0.0%
μ_T	TB disease specific mortality rate	9.8%	2.9%
k	Self-cure rate	0.4%	1.1%
ECR	Effective contact rate	23.6%	1.2%
i	Proportion progress to infectious disease	1.5%	0.3%
p	Proportion progress to primary disease	34.3%	3.5%
v	Endogenous reactivation rate	0.3%	0.0%
γ	Susceptibility to reinfection	1.7%	0.6%
f	Resistance transmission reduced by	6.9%	45.9%
w	Relapse rate	12.5%	1.2%
Treatment parameters			
nr	Notification rate	-----	-----
Φ_S	Treatment success rate for DS cases	-----	-----
Φ_R	Treatment success rate for DR cases	-----	-----
r	Acquired resistance rate	0.1%	44.1%

D.





Scenario 8. *Model sensitivity under a poor control program.* The results of 1000 Monte Carlo simulations run for ten years under no treatment (notification rate set to 0) are shown as the final incidence of infectious disease (A). Model parameters were randomly generated in each simulation, assumed to have a normal distribution with mean and variance as given in (B). The Pearson squared correlation (r^2) calculated in (C) reflects the proportion of variance in model outcome explained by variance in each parameter. Scatterplots for the input parameters which most influenced the model outcome of resistance prevalence are shown in (D).

14. Appendix 4

Drugs used in chemotherapy

14. Appendix 4
Drugs used in chemotherapy
I. Isoniazid
I.A. Bacterial targets
I.B. General effects: bacteriostatic and bacteriocidal
I.C. Pharmacokinetics
I.D. *In vivo*: human treatment
I.E. Emergence of resistance
I.F. Summary
II. Rifampicin
II.A. Mode of Action
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III.B. In vitro studies
III.C. Dosing
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I. Isoniazid

Isonicotinic acid hydrazide, or isoniazid (INH), is one of the most commonly used drugs for the prophylaxis and treatment of mycobacterial diseases. (Rattledge 1999) Isoniaizd is selective for mycobacteria. (Goodman 1996) *M. tuberculosis* and *M. bovis* are both very susceptible to INH, with MICs ranging from 0.02 to 0.2 ug/ml. (Rattledge 1999) Other mycobacterial species are less susceptible, with MICs of 5 to 50 ug/ml. (Pansy 1952) Non-mycobacterial bacteria are fairly resistant to INH action. *E. coli*, for example, is resistant to at least 500 ug/ ml. (Rattledge 1999)

I.A. Bacterial targets

Isoniazid is thought to exert a bactericidal action on mycobacteria by inhibiting synthesis of mycolic acids, a component of the mycobacterial cell envelope. INH has been shown to inhibit mycolate synthesis in vitro within six hours, at levels of 0.1 ug/ml (equal to the MIC for *M. tuberculosis*) for human strain H37Ra and 0.5 ug/ml for *M. bovis*-BCG. (Takayama, Wang et al. 1972) *In vivo* evidence has also implicated mycolic acid synthesis as a downstream effect of isoniazid on tubercle bacilli. The rate of mycolic acid synthesis inhibition has been shown to increase with the rate of uptake of INH.(Wang and Takayama 1972) The inhibitory action of isoniazid has been shown to be reversible, if washed out before 10 hours. (Takayama, Wang et al. 1972)

The strong specificity that isoniazid shows for mycobacterial species was explained with the discovery of the drug's cellular target. Mycolic acid is a cell wall component of all mycobacterial species. The considerable variability of MICs amongst mycobacterial species is generally accounted for by differential cell wall permeability to isoniazid. (Rom 1996) Recent studies have also explained this difference by the lack of a functional *oxyR* gene in *M. tuberculosis*. *OxyR* encodes a transcriptional activator of *aphC* transcription, which itself encodes a subunit of alkyl hydroperoxide reductase. AphC and oxyR are both stimulated in response to oxidative stress. Mutations in the promoter of the aphC gene have appeared as compensatory mutations in INH-resistant bacilli with a mutation in the *katG* gene. *KatG* encodes a protein with catalase-peroxidase activity. The overexpression of the *aphC* hydroperoxidase is thought to compensate for this reduced activity in *katG* mutants. (Rattledge 1999)

I.B. General effects: bacteriostatic and bacteriocidal

INH can exert both a bacteriostatic and bactericidal activity on tubercle bacilli. The bactericidal action is generally exerted on growing cells, and the bacteriostatic action on resting cells. (Goodman 1996) Isoniazid kills cells in lag or logarithmic, but not stationary phase. In the first 24 hours after exposure to the drug, only young cells are killed. (Barry 1964) The tubercle bacilli generally undergo 1-2 divisions after treatment with isoniazid, before cellular multiplication is arrested. (Goodman 1996) The bactericidal action of INH is almost independent of concentration, at doses above 0.1 ug/ml. (Mackaness and Smith 1953; Barry 1964)

M. tuberculosis strains exposed to isoniazid show a period of delay between the time of exposure and the observation of a bactericidal effect. The length of this delay

period depends on bacterial growth rate, but is largely independent of drug concentration. There is usually a delay of 1 – 4 days before bactericidal action is seen, corresponding to about 1 ½ generations. (Mackaness and Smith 1953; Barry 1964)

INH penetrates easily penetrates into sensitive cells. (Goodman 1996) Thus, the drug is equally effective for intracellular and extracellular bacilli. (Goodman 1996) The mechanism of cellular uptake is believed to involve active transport. (Gangardharam 1997) The process of INH uptake has been shown to require oxygen (uptake is inhibited under anaerobic conditions) and is largely inhibited by heating. (Gangardharam 1997) Not surprisingly, INH has also been shown to be inactive in anaerobic conditions. (Barry 1964)

Tubercle bacilli treated with INH undergo morphological changes. The bacilli turn in shape to appear as tapering rods. The internal cellular structure lost within 24 hours. (Barry 1964) INH causes the loss of acid-fast property of mycobacteria, as the cell wall becomes permeable to the decolorizing agent used in the test. (Rattledge 1999) This occurs at same time as the onset of bactericidal action (one to two generations). (Barry 1964)

I.C. Pharmacokinetics

1. *In vitro* studies

The minimal tuberculostatic concentration *in vitro* is 0.025 to 0.05 ug/ml. (Goodman 1996) The concentration needed to inhibit the tubercle bacilli multiplication is little affected by the size of the inoculum, suggesting that only a small proportion of the drug is adsorbed onto cells. (Barclay 1964; Barry 1964)

I.D. *In vivo*: human treatment

The small molecular size of INH allows wide distribution, and INH diffuses readily into all body fluids and tissues. (Chamgers 1998) Isoniazid can be found detected in serous membranes, caseous foci, tubercle cavities, and macrophages. (Dannenberg 1998)

The metabolism of INH is genetically determined, and depends on the rate of acetylation of the drug. (Chamgers 1998) INH is metabolised in the liver by both acetylation and oxidation. (Dannenberg 1998) For rapid acetylators, the average concentration of INH in plasma is about 1/3 to ½ of that in slow acetylators and the average half-lives are less than 1 hr and 3 hrs, respectively. The more rapid clearance

rate by rapid acetylators is an important consideration in therapy if the drug is not taken on a daily basis. Following an average adult oral dose of 5 mg/kg (300 mg), peak serum concentrations of INH reach 3 – 5 ug/ml in 1- 2 hours. (Chamgers 1998; Dannenberg 1998) The inactive metabolites of INH are excreted in the urine. Approximately 75 – 95% of a dose is removed within 24 hours. (Dannenberg 1998)

Studies have suggested a two-phase action for INH, consisting of: 1) a reversible uptake in sensitive cells (whether actively growing or not), and 2) an irreversible binding (occurring only in growing cells). (Barry 1964) In the first phase, isoniazid uptake by sensitive growing bacilli occurs easily, peaking one day after exposure. The rate of uptake then falls, and subsequently slowly increases to a plateau level at 7 days, by which time the bactericidal action has been completed. (Barry 1964) Resistant strains show much lower ability to uptake isoniazid, suggesting the failure to accumulate drug as a mechanism of resistance. In the second phase of INH action, the binding of INH to cells is irreversible; a 10 day incubation in drug free medium does not reverse INH action on cells. (Barry 1964)

I.E. Emergence of resistance

M. tuberculosis shows a mutation rate to isoniazid of $1 - 3 \times 10^{-6}$ / bacterium / generation. The endogenous frequency of mutants resistant to 0.2 – 5 ug/ml INH, observable in a sensitive population, is $1/10^5$ to $1/10^7$. Such resistance mutants lack the catalase enzyme, occur in single step mutation, and are considered high resistance (growing on 20 ug/ml or greater of INH) mutants. (Barry 1964)

The current hypothesis for the molecular basis of isoniazid action is that the drug is oxidized by catalase-peroxidase (encoded by the *katG* gene) to an active compound, which binds to the *inhA* gene product (an enoyl-ACP reductase). (Rattledge 1999) Resistance to INH action arises in two main fashions: either mutations in the *katG* gene prevent conversion of INH to the active form of the drug, or mutations in the structural gene *inhA* prevent its action. (Rattledge 1999) Low level of resistance is seen in mutants that overexpress the *inhA* gene product. Highly resistant isolates usually show a missense or complete deletion of the *katG* gene. (Dannenberg 1998)

Most commonly, resistance is due to a change in the *katG* gene; 50 to 75% of INH-resistant isolates have a *katG* missense mutation or deletion. (Rattledge 1999) The role of *katG* mutation in causing resistance to INH has been further verified by studies showing that the insertion of a wild type copy of *katG* into an INH resistant mutant with

a *katG* mutation does restore drug sensitivity. (Gangardharam 1997) Sequence analysis of *katG* mutants resistant to INH has showed the development of a compensatory mutation in the promoter region of the *aphC* gene. Such a compensatory mutation is likely to counteract the loss of catalase/peroxidase activity by hyperexpressing the alkyl hydoperoxidase encoded by *ahpC*. (Rattledge 1999) Strains producing catalase are more virulent then *katG*-deficient mutants in immunocompetent mice. (Rattledge 1999)

Two factors—INH uptake and enzyme activity—have been shown important in resistant mutants. One category of resistant mutants clearly shows a deficiency in their ability to take up INH from the environment. Isoniazid resistant cells exposed to ^{14}C-INH have taken up much less radioactivity than corresponding sensitive cells. (Gangardharam 1997) Studies have also shown that in mutants with a high level of resistance to INH, the primary mechanism of resistance is the decreased permeability to the drug. The resistant mutants appear to have acquired a cell wall factor impermeable to isoniazid. (Gangardharam 1997)

I.F. Summary

Isoniazid is one of the leading anti-mycobacterial drugs used in the treatment of tuberculosis. Experimental studies, both *in vitro* and *in vivo*, have elucidated the mechanisms and dynamics of INH action on mycobacteria. Genetic studies on *M. tuberculosis* have suggested the genetic basis of resistance to INH action, as well as identifying the molecular drug targets. This understanding of INH drug action on *M. tuberculosis* provides us important information in modelling the interaction between the drug and the bacteria. In previous sections we have provided initial models of the drug treatment of a tuberculosis infection. In the following section we add upon these basic models of drug treatment, incorporating the specifics from this literature review of isoniazid action.

II. Rifampicin

II.A. Mode of Action

Rifampin is a large (MW 823) complex that serves as an antituberculosis drug. Rifampin is in the class of rimacyins, a group of structurally similar, highly lipophilic, antibiotics. Rifampin is a semi-synthetic derivative of rifamycin B. RIF inhibits most gram positive, as well as many gram negative bacteria. Is active equally well on mycobacterial and nonmycobacterial organisms. Because of its high versatility,

rifampin is also used to treat other mycobacterial diseases, including leprosy. (Rattledge 1999) (Goodman 1996; Gangardharam 1997; Chamgers 1998)

RIF acts by inhibiting RNA polymerase enzymatic activity and thus blocking RNA synthesis. Rifampin binds directly to the DNA-dependent RNA polymerase, forming a stable complex. The drug acts on the process of transcription, prior to the formation of the first phosphodiester bond. (Gangardharam 1997) The maximal levels of inhibition are achieved in vitro assays by incubating the drug and enzyme prior to the addition of other necessary assay components. (Gangardharam 1997) RNA polymerase from M. tuberculosis is 1000X more sensitive then from E. coli. (Gangardharam 1997)

II.B. Bactericidal activity

Rifampin is bactericidal for mycobacteria. (Goodman 1996) RIF readily penetrates most tissues and into phagocytic cells, easily diffuses across the hydrophobic mycobacterial cell envelope, and is bactericidal for intracellular organisms. (Chamgers 1998) Rifampin is considered a sterilizing agent in the chemotherapy of tuberculosis, in that it is active against slowly metabolising organisms. (Dannenberg 1998)

II.C. Mechanism

RNA polymerase consists of two parts: the core enzyme, (which has four subunits, including one called beta), catalyses transcription elongation. The holoenzyme includes the core and sigma factors that show specific affinity for different promoters, and function at the level of transcription initiation. (Rattledge 1999) Rifampicin binds the beta subunit, inhibiting transcription. (Goodman 1996; Gangardharam 1997)

Rifampin exerts its bactericidal effect on cells by inhibiting the production of RNA synthesis. Rifampin binds strongly to the beta subunit of the bacterial DNA-dependent RNA polymerase, forming a stable drug-enzyme complex. (Chamgers 1998) Rifampin requires the presence of the entire RNA polymerase complex in order to bind; but the drug site of action is the beta subunit. The formation of this complex suppresses the initiation of chain formation of RNA synthesis. Rifampin is unable to bind to the nuclear RNA polymerase found in eukaryotic cells, and therefore does not inhibit RNA synthesis in these cells. (Goodman 1996)

II.D. In human action/ dosing

The growth of *M. tuberculosis* is inhibited in vitro by 0.005 to 0.2 ug/ml RIF. (Goodman 1996) For myobacterial infections, RIF used 600 mg/d/ (10 mg/kg/d) orally. (Goodman 1996) In some short-course therapies, use 600 mg given twice weekly. Rifampicin is well absorbed after oral administration, excreted mainly through the liver into bile. Usual doses result in serum levels of 5-7 ug/mL. Rifampicin is widely distributed in body fluids and tissues and is found to be highly protein-bound. (Chamgers 1998) Rifampin penetrates into tissue, caseous foci, serous membranes, and macrophages. (Dannenberg 1998) Oral dosing gives peak concentrations in plasma 2-4 hours. After 600 mg, peak serum concentration is 7 ug/ml, but there is much variability amongst patients. The half-life varies from 1.5 to 5 hours, longer in case of hepatic dysfunction. (Goodman 1996) Half life progressively shortened by 40% during first 14 days of treatment; induction of hepatic microsomal enzymes with acceleration of drug deacetylation. (Goodman 1996)

Rifampin is metabolised in the liver and excreted in the bile. Rifampin can be detected in body fluids including urine, tears, and sweat, colouring them orange-red. (Dannenberg 1998) Use of rifampicin shows a low incidence of bad side effects, and is generally well tolerated, with less then 4% adverse reactions. (Goodman 1996)

II.E. Emergence of resistance

Resistant mutants are present in all microbial populations at frequency of about 1 in 10^7 to 10^8 bacilli. Resistance results from one of several possible point mutations, which prevent RIF binding to RNA polymerase. Resistance is seen to develop rapidly in vitro in a one step process. As the human RNA polymerase is not able to bind to rifampin, it is also unable to be inhibited by the drug. (Chamgers 1998) (Goodman 1996)

The gene coding for the beta subunit of RNA polymerase is rpoB. Mutations in this gene give a high level resistance to rifampicin. Over 90% of rifampicin resistant bacilli have a mutation in rpoB. These are typically missense point mutations occurring in a region of 27 codons near the centre of the gene. These mutations do not appear to confer any significant growth disadvantage. 5% of clinically isolated resistant strains do not have a mutation in rpoB, suggesting that there exists an alternative mechanism of resistance. A likely possibility is thought to be the inactivation of rifampicin, as is seen in other species of mycobacteria. RIF inhibits uridine-C14 uptake in sensitive, but not in

resistant strains. Mutants that have the rifampicin mutation in both the alpha and beta subunits are resistant overall; bacteria which have only the beta subunit with a resistant mutation still show resistance to drug action. (Aboshkiwa, al-Ani et al. 1992; Telenti, Imboden et al. 1993; Aboshkiwa, Rowland et al. 1995; Gangardharam 1997; Telenti, Honore et al. 1997; Rattledge 1999; Morlock, Plikaytis et al. 2000)

Most of all isolated rifampicin resistant mutants show mutation located in rpoB gene, in a 69bp region. This small region in rpoB has a higher potency for mutation to resistance. Most of clinically isolated resistant mutants have shown mutations in a short region of 27 codons in the center of the rpoB coding sequence. These mutations are mostly missense mutations, although some small in-frame insertions and deletions have also been found. (Honore and Cole 1993; Telenti, Imboden et al. 1993; Gangardharam 1997; Telenti, Honore et al. 1997)

III. Streptomycin

III.A. Mode of Action

Streptomycin, a member of the aminoglycoside family of antimicrobial agents, exerts a bacteriocidal action on mycobacteria. Streptomycin is specific to M. tuberculosis, and non-tuberculous mycobacteria are resistant to its action. The drug acts by interfering with protein synthesis, targeting the bacterial ribosome. (Goodman 1996) The bactericidal action of streptomycin is exerted only on growing organisms. The completeness of the killing action appears proportional to the rapidity of replication. (Barry 1964) Streptomycin action is related to the size of the inoculum. The proportion of SM bound to bacteria is usually small compared to the amount free in solution. (Barry 1964)

After a dose of streptomycin near the MIC (1 to 10 ug/ml for M. tuberculosis), the tubercle bacilli can continue to grow for several generations. This is referred to as a delay period, in which growth continues until the bactericidal effect of the drug commences. During this delay period, for about two days after treatment with streptomycin, the bactericidal action of the drug can be prevented by washing off the drug. After this time, bacterial lysis begins to take place, reducing the bacterial number to about the level at the start of treatment. While during this delay period the number of bacteria continues to increase, the number of viable units decreases. After the induction of bactericidal activity, the number of viable units continues to decrease. At

higher dosages of streptomycin, at concentrations of over 10 ug/ml, the progression to bactericidal action is much more rapid. Once the drug is administered to tubercle bacilli at such a dose, the effects are non-reversible, and will result in the death of the bacilli. (Barry 1964)

Streptomycin shows both bacteriostatic and bacteriocidal activity. The determination of which activity predominates in any given interaction with a bacterium is related to drug concentration used, contact period, targeted organism, growth rate, medium used, and inoculum size. For the treatment of M. tuberculosis in human patients, the bactericidal action of streptomycin usually prevails. (Barry 1964)

Tubercle bacilli treated with streptomycin show morphological changes. The immediate change is the elongation and swelling of the bacteria. The secondary morphological change, seen after four days exposure, is the loss of cytoplasmic structure and the disintegration of the cytoplasm. (Barry 1964)

III.B. In vitro studies

Streptomycin, because of its highly polar nature, penetrates into cells poorly. The drug is therefore primarily active against extracellular tubercle bacilli. (Chamgers 1998) Studies have shown poor action against intracellular bacilli. (Chamgers 1998)

In vitro studies have shown that relatively low concentrations (on the order of 2.5 x 10^{-9} M) of streptomycin can inhibit protein synthesis. And in theory, only one molecule of SM per ribosome is needed to inhibit the protein synthesis of a cell. (Gangardharam 1997) However, a larger dose is needed for effective bactericidal action against bacterium such as M. tuberculosis, for which there exist barriers to cellular uptake of the drug. Excess concentrations must be used in such cases, to account for the relative permeability of the targeted cells. (Gangardharam 1997) Most tubercle bacilli are inhibited by a dose of 1 – 10 ug/ml in vitro. (Chamgers 1998) A sensitive population of M. tuberculosis can be expected to be about 80% inhibited by such a dose of streptomycin. (Gangardharam 1997)

III.C. Dosing

Streptomycin is administered by intermittent intracellular injections. For the treatment of tuberculosis, SM is usually given at a dose of 15 mg/kg per day with a single intramuscular injection. (Goodman 1996)

An intramuscular injection is absorbed well, giving peak blood concentrations within 30 –90 minutes. Streptomycin is usually given in human therapy intravenously, as a 30 – 60 minute infusion. Such treatment yields a brief distribution phase, followed by serum concentrations identical to those seen after an intramuscular injection. Therapy is usually given in 1 to 3 equal daily doses (for patients with normal renal function). (Chamgers 1998)

The dose of streptomycin is cleared by the kidney. After an oral dose, virtually entire dose is excreted in feces. The drug excretion rate is directly proportionate to creatinine clearance. The normal half-life of the drug in serum is 2-3 hours, increasing to 24-28 hrs in patients w/ significant impaired renal fxn. (Chamgers 1998)

III.D. Emergence of resistance

On average, 1 in 10^8 tubercle bacilli can be expected to show natural resistance to streptomycin, growing on media containing 10 – 100 ug/mL streptomycin. (Chamgers 1998) The most common mechanism of streptomycin resistance is mutation of genes affecting the bacterial ribosome, which prevent the enzymatic inhibitory action of streptomycin. The primary mutation found in human tuberculosis cases resistant to streptomycin is a point mutation that alters the ribosomal binding site. (Meier, Kirschner et al. 1994; Chamgers 1998) Other types of resistant mutants are found, including those resulting from the acquisition of enzymes that can metabolize or prevent the up-take of streptomycin. (Goodman 1996)

Three categories of single step mutants are typically observed in a sensitive population: low resistance (at a frequency of 10^{-6}), medium resistance (10^{-7}), and high resistance (10^{-9}). (Barry 1964) One also observes resistance emerging through a series of steps in succession. The frequencies for such multiple-step mutants vary greatly by strain. Spontaneous mutation of tubercle bacilli to a high level of resistance to streptomycin has been observed at a rate of about 10^{-10} / bacterium. (Barry 1964)

Mutants showing a high level of resistance to steptomycin require a 10,000 fold higher concentration then used for sensitive strains, to achieve a 50% inhibition. (Gangardharam 1997) The mutations conferring such high levels of resistance have been found to reside in the genes for the bacterial ribosome. In contrast, *M. tuberculosis* mutants showing low levels of resistance appear to use an alternative mechanism for resistance. Such mutants show similar in vitro inhibition of protein synthesis, showing

that the mutation conferring cellular resistance to SM does not reside in ribosome function. (Gangardharam 1997)

DRUG	CELL TARGET	INHIBITS	Requires Growth?	ACTIVE AGAINST	Environ -ment	Sterilizing Activity in vivo?	Mechanism Resistance	Mutation Rate	Endemic Resistance
FIRST LINE DRUGS									
ISONIAZID (INH)	Enzyme required for mycolic acid synthesis	Cell wall synthesis	Yes	Rapidly dividing; weakly on slowly multiplying	Extracellular	None	Inactivation of catalase-peroxidase gene, katG	10-8	1 in 106
RIFAMPICIN (RMP)	B- subunit RNA polymerase	RNA synthesis	Yes	acts against slowly (+ rapidly) growing organisms	Intra + Extracellular	Sterilizing (acts on persisters)	RpoB mutation (β-subunit pol)	10-10	1 in 108
STREPTOMYCIN (SM)	ribosome	Protein synthesis	Yes	Rapidly dividing	Extracellular	Slightly Sterilizing		10-8	1 in 107
PYRAZINAMIDE (PZA)	?	?	Bacteriostatic	Can kill in acid environment	Intra + extracellular	Sterilizing		10-3	1 in 106
ETHAMBUTOL (EMB)	Arabinogalactan synthesis	Cell wall synthesis	Yes	BACTERIO-STATIC	Weakly active against Intra + Extracellular bacilli	None	Mutation of target enzyme	10-7	1 in 105
SECOND LINE DRUGS									
ETHIONAMIDE	Inhibits mycolic acid			BACTERIO-STATIC			Mutation of target	10-3	

	synthesis						enzyme		
Thiacetazone				BACTERIO-STATIC				10-3	
P-aminosalicyclic acid (PAS)		Folate biosynthesis		BACTERIO-STATIC				10-8	
Oxfloxacin		DNA gyrase		Rapidly dividing; Neutral or alkaline pH			gyr A&B	?	
QUINOLONE	DNA Gyrase	DNA replication					Mutation of target enzyme	?	

Table 14-1. The properties of drugs used in the treatment of tuberculosis. Sources: (Mackaness and Smith 1953; Mitchison and Fox 1969; Mitchison 1974; Mitchison 1979; Goodman 1996; Gangardharam and Jenkins 1997; Mitchison 1997; Peloquin, Jaresko et al. 1997; Mitchison 2000)

DRUG	Dose	Cmax (mg/L)	Tmax	$t_{1/9}$	MIC	Protein binding	Inhibits	Requires growth?	Active against
ISONIAZID	300	5	1.0 (Peloquin, Jaresko et al. 1997)	3	0.1	0	Cell wall synthesis	Yes	Rapidly dividing; weakly on slowly multiplying
(slow acetylator)									
(fast acetylator)		4		1.3					
RIFAMPICIN	600	12	1-2 (Peloquin, Jaresko et al. 1997)	3	0.3	85	RNA synthesis	Yes	acts against slowly (+ rapidly) growing organisms
STREPTOMYCIN	750	40		3-5	2	35	Protein synthesis	Yes	Rapidly dividing
PYRAZINAMIDE	2000	40		8	20	0	?	Bacteriostatic	Can kill in acid environment
ETHAMBUTOL	1200	3		3	1.5	0		Yes	Low activity

Table 14-9. The pharmacokinetic and pharmacodynamic properties of drugs used in the treatment of tuberculosis. Sources: (Mackaness and Smith 1953; Mitchison and Fox 1969; Mitchison 1974; Mitchison 1979; Goodman 1996; Gangardharam and Jenkins 1997; Mitchison 1997; Peloquin, Jaresko et al. 1997; Mitchison 9000)

15. Appendix 5

Estimation of within-host mycobacterial growth rate under drug treatment

15. Appendix 5
Estimation of within-host mycobacterial growth rate under drug treatment
I. Introduction
II. The Data Set
III. Methods of fitting model to data
IV. Estimating *in vivo* growth of TB strains
V. Estimating *r*
VI. Discussion

I. Introduction

We consider here a model of a tuberculosis infection in order to better understand the dynamics of the within-host mycobacterial population.

In particular, we are interested in quantifying the *in vivo* growth rate of *Mycobacterium tuberculosis*, as this directly affects the rate of progression to infectious tuberculosis disease, as well as the time needed to treat and clear an infection and the rate of emergence of drug resistance. Each of these points is crucial to understanding current issues in the treatment of tuberculosis, in particular the epidemics of drug resistance and the need for a long duration of treatment.

We know from *in vitro* experiments that mycobacteria tuberculosis is slow growing, dividing on the order of once per 20-24 hour period, depending on conditions used. (Fishman and Elias 1998; Colston 1999) However, it has been difficult to infer from these experiments an *in vivo* growth rate, as the environment within-host is difficult to characterize and replicate in an *in vitro* setting.

In particular, the multi-compartmental nature of a mycobacterial population within an infected tuberculosis patient, including sub-populations that may have differential access to nutrients as well as drug, confound these interpretations. In addition, the host immune system plays a role in suppressing mycobacterial growth, limiting the population size.

While it is widely accepted that mycobacteria grow more slowly within-host than in optimal *in vitro* conditions, experiments have not been able to disentangle host effects on bacterial death from inhibition (or slowing) of growth. In the terms of population dynamics, this means we cannot differentiate between an increased death rate and a decreased division rate. However, we can comment on the relationship between the birth and death rate. In particular, we can estimate the bacterial growth rate, defined as the birth rate divided by the death rate.

Previously, the *in vivo* growth rate of mycobacteria remained fairly elusive due to limited quantitative bacteriologic data from tuberculosis patients. Recent studies measuring the colony forming units (CFU) of mycobacteria over the course of patient treatment provide us the opportunity to estimate the growth rate. (Nunn 1992; Brindle 1993; Desjardin 1999)

We examine previously reported data on the timecourse of CFU during the first 28 days of treatment of infectious tuberculosis patients with a daily regimen of streptomycin (S), thiacetazone (T), and isoniazid (H). (Brindle 1993) We propose a model to explain the decrease in mycobacterial CFU over this treatment time.

In particular, we propose the existence of two distinct sub-populations of mycobacteria within the host, which differ only in their effective growth rates.

Further, we add to previous population dynamic models of bacterial infection by including specific pharmacodynamic parameters to account for the mode of drug action on bacteria. As the drugs used in this treatment (S, T, and H) each require growth to exert bactericidal activity, we suggest that the drug-induced death rate is proportional to the mycobacterial growth rate.

We estimate these bacterial population dynamic parameters by fitting our model to the described data set, using maximum likelihood. We find that our model of two bacterial populations with equal drug susceptibility but different growth rates can explain the observed data and variance with a high level of significance ($p<0.001$), and suggest interpretations of the parameters as well as alternatively plausible models.

II. The Data Set

We examine the counts of mycobacteria present in sputum of patients undergoing the first month of a standard regimen of tuberculosis treatment. This data was part of a study to compare the chemotherapeutic effect of two particular TB regimens in HIV-positive and a control HIV-negative patient group, in Nairobi, Kenya.

All patients in the study were confirmed TB positive, had not received previous TB treatment, and showed no initial drug resistance to the drugs used. Two different TB regimens were compared in this study—STH and SHRZ (streptomycin, isoniazid, rifampicin, and pyrazinamide)—in both HIV positive and negative patient groups. (Brindle 1993)

Brindle compares these treatment regimens and concludes that HIV status does not significantly change the effectiveness of chemotherapy, as measured by the rate of decrease of mycobacterial counts in the sputum. This suggests that drug bactericidal activity is similar in HIV-positive and negative patients, even though the difference in immune system response to the mycobacteria must differ significantly. (Brindle 1993)

In the second phase of this study, patients were given a continuation treatment regimen (of TH) for an additional 11months and mortality rates and causes of death in the four treatment groups were compared. (Nunn 1992)

Mitchison's statistical analysis of the Nairobi data suggested experimental and biological explanations for the variance seen in patient drug response. (Mitchison, in press).

We focus on the patients receiving STH treatment, as we are interested in understanding the effects of bactericidal drugs requiring growth. The second treatment includes two drugs—rifampicin and pyrazinamide—that are considered 'sterilizing' drugs and are shown *in vitro* to be active on slow growing mycobacteria.

The patients are given daily doses of streptomycin, thiacetazone, and isoniazid. Sputum samples are taken at unequal intervals during a 28-day period after the beginning of treatment. Mycobacterial numbers present in sputum are measured by colony counts, and reported as $\log_{10}$counts.

III. Methods of fitting model to data

We tested the distribution of CFU counts for normality, using SPSS K-Wallis test. We found that the bacterial counts did not significantly conform to the normal distribution. In addition, we found that the variation changed with time. We transformed the data in order to work with a normal distribution and to normalize the variation.

We found that the mean of the CFU counts is approximately equal to the standard deviation. As this is a property of the log normal distribution, we considered transforming the data by taking the natural logarithm of the counts. We find that the LN

(counts) for timepoints 2 to 28 does indeed more significantly (by the K-Wallis test) resemble a normal distribution. However, the counts at timepoint t=0 did not conform to normality.

In order to get around the high variance present during the rapid kill phase of the first two days of treatment, we examined the relative kill in each patient. We plotted the relative LN(counts) at each time point, defined as the LN N(t) / LN N(0). We found that the relative LN counts did significantly fit a normal distribution.

We fit our model (see above) to this data set, and estimated parameters by maximum likelihood. We find, for the log normal distribution, that the variance (σ^2 = 0.035) is constant over time. We use the sample estimate of the variance from the patient data set as the MLE of the variance in our model.

We find the MLE for our model parameters by minimizing the negative log likelihood for a normal distribution, calculated as:

$$\text{- Log likelihood} = n \log(\sigma) + \frac{n}{2} \log(2\Pi) + \frac{(\mu - \overline{\mu})^2}{2\sigma^2}$$

We minimize this function and fit parameters with a C++ program using powell minimization. We found powell to be more efficient and reliable method than Newton or conjugate minimization.

We found confidence intervals for each parameter by refitting the model at each point, holding the parameter of interest constant. The 95% confidence interval is found for maximum likelihood estimates that fall within a x^2 with 1 d.f., reflecting the one parameter that is held fixed.

$$\chi^2_{1df} = 3.84$$

Change in log likelihood < 3.84

A change in log likelihood less than 3.84 falls within the 95% confidence interval. We found the parameter confidence intervals using powell minimization in C++, through repetitive calls to minimize the negative log likelihood function as one parameter was given a fixed value and the remainder were allowed to vary.

IV. Estimating *in vivo* growth of TB strains

We are interested in quantifying population parameters that describe the within-host mycobacterial growth rates, as a template to measure drug action. For viruses, the convention is a parameter known as R_0, which describes the number of secondary cell infections by a primary infected cell, in the absence of constraints on viral population

growth. This parameter is useful in describing populations growing primarily intracellularly, and as such, is less applicable to studies of active infectious tuberculosis.

We consider an environment in which mycobacteria are at equilibrium with any host-induced growth restraints, particularly immune response. In addition, we focus our attention on the mycobacterial population that grows extracellular. While early after infection the mycobacterium is primarily growing within macrophages, by the time there has been progression to active, smear positive tuberculosis disease, the mycobacteria are primarily extracellular. The liquefied caseous material found in the now aerated foci seem to inhibit macrophages, and for the first time since the start of infection, the tubercle grows in an extracellular environment.

We therefore assume that the mycobacterial population grows extracellularly. A relevant parameter to describe the extracellular growth of a bacterial population is the intrinsic population growth, r, which describes the potential for growth in the absence of constraints.

We measure here the intrinsic population growth in the presence of specific-activated host immune response. We also measure the intrinsic growth rate in the presence of drug, during the first 28 days after treatment. The drug acts, as expected, to bring the intrinsic growth below 0, ($r<0$), such that the drug reduces the mycobacterial population size, already found at equilibrium with the host immune response.

V. Estimating *r*

We model two bacterial strains growing exponentially and independently. We assume here that the growth rates are density-independent and do not attempt to apply an upper limit to the population size. We feel these assumptions are appropriate for this model, as we are considering a bacterial population that begins at t = 0 at drug-free equilibrium. We then introduce drug treatment and observe the change in population size during short intervals. During this time, the bacterial population can be assumed to be adjusting to the addition of drug and (far from equilibrium) undergoing exponential decline. We assume constant drug concentration over time.

$$\text{Model 1: } N_t = N_1 e^{r^*_1 t} + N_2 e^{r^*_2 t}$$

We first estimate the intrinsic mycobacterial growth rate in the presence of drug, r^*. We consider a model in which two mycobacterial strains are present. We estimate the growth rate of each strain, and find that the two growth rates are distinct ($r^*_1 \neq r^*_2$)

with $p<0.05$. We estimate the death rate of strain 1 to be 0.11 (95% CI [0.06, 0.14]) that of strain 2. We estimate that 9% of the total mycobacterial population is being killed at this slower rate.

We fit model 1 to the data set using maximum likelihood estimation, and found that the maximum likelihood estimate for model 1 was 263.28. To test whether this model can significantly represent the data, we compared the ML (itself a chi-squared distribution) to a χ^2 with 4 d.f.. We found that this model can predict the data with a p-value of $p<0.01$.

Our estimation of the growth rate of two strains during this treatment suggests that approximately one in ten mycobacteria is cleared at a rate of 6 – 14% of the remaining mycobacteria. This estimate of r^* represents the mycobacterial growth rate *in vivo* in the presence of drug treatment. This reflects a combination of the intrinsic mycobacterial growth rate (births), the death (or decreased birth) rate due to host immune response, and the death rate from drug action.

$$r = births - deaths$$

We then seek to understand the two components of the growth rate, the birth and death rates. We propose three models for the combination of birth and deaths that can explain the dynamics for the clearance of the two mycobacterial populations under drug treatment. We find that each of these models (models 2 – 4) can predict the variance seen in the data with greater than 99% confidence, but that none is significantly better explanation of the data ($p<0.01$).

In models two and three, we present two strains which share either a common birth or death rate. In model four we consider a situation where the death rate is directly proportional to the birth rate, and this proportionality constant is shared between two strains with distinct birth and death rates.

Model 2: $N_t = N_1 e^{(\mu - D_1)t} + N_2 e^{(\mu - D_2)t}$

We model here two strains (N_1 and N_2) growing with a birth rate (μ) but distinct death rates (D_1 and D_2). This is meant to represent bacterial strains that either have differential drug susceptibility or drug access. We estimate that the initial population consists of 91.1% strain 1 and 9.9% strain 2. We estimate the birth rate μ as 0.91 divisions/ day, D_1 as 1.09 deaths/ day, and D_2 as 1.99 deaths/ day. We then estimate the

dimensionless ratio births / deaths, representing the strains growth capacity. We find the best estimate for μ/D_1 to be 0.84, approximately twice the best estimate of 0.39 for μ/D_2.

Model 3: $N_t = N_1 e^{(\mu_1 - D)t} + N_2 e^{(\mu_2 - D)t}$

We model here two strains (N_1 and N_2) with distinct birth rates (μ_1 and μ_2) but a common death rate (D). This represents strains growing at different rates due to either differential access to nutrients required for growth or the extent of immune inhibition.

Model 4: $N_t = N_1 e^{\mu_1 (1-d)t} + N_2 e^{\mu_2 (1-d)t}$

where $D = d\,\mu$

We model here two strains (N1 and N2) with both distinct birth rates (μ1 and μ2) and death rates (D1 and D2). We assume that the drug-kill rate is directly proportional to the bacterial growth rate. D is the proportionality constant for drug action on the dividing bacteria. This relationship allows us to model drug action that is dependent on the growth rate, requiring division in order to kill the bacteria. Organisms that do not divide ($\mu = 0$) will not be killed by drug (drug-kill rate = 0). The faster a bacterial strain divides, the faster it will be killed by drug.

This model incorporates our intended requirement for bacterial division in order for the drug to exert its bacteriocidal activity. In addition, we assume here that all bacteria are killed with the same rate per cell division. The difference seen in growth rates between the two bacterial strains is due solely to the relative rates of division.

Drug – kill rate $= d\ \mu$

Drug-induced deaths $= N_0 e^{d\,\mu\,t}$

If D = 1, then birth rate = death rate, and strain size will stay constant.
If D>1, then drug will decrease the population size If D<1, then growth rate will exceed drug-death rate, and population will grow

We estimate the initial population to consist of 5% strain 1, with a birth rate (μ_1) of 0.13 divisions/day. Strain 2 was estimated at 95% of the initial population, with a birth rate (μ_2) of 1.2 births/day. The common drug death rate constant (d) was estimated as 1.99. The drug death rate ($D = d\,\mu$) for strain 1 (D_1) was 0.08-0.51 deaths/day, and for strain 2 (D_2) 1.27-4.00 deaths/day. We also estimate the birth / death ratio as 0.41 – 0.63, a narrower interval than we obtained in the above models.

We find that these three proposed models of bacterial growth rates are equally likely and thus we cannot reject any of these models. If we made assumptions about the relation between the birth and death rates, including the extent of variation within the bacterial population and the ratio between birth / deaths, we could estimate these rates as well. However, a better biological understanding is required to distinguish which assumptions are valid and which model is most applicable.

VI. Discussion

While the nature of dormant or persisting mycobacteria remains elusive, the existence of slow growing strains able to evade the host immune response is believed to be an important component in treatment failure. Our within-host model of the dynamics of TB growth predicts that one strain is cleared at a significantly slower rate, reinforcing this notion.

M. tuberculosis has the ability to remain dormant for extended periods of time. (Colston 1999; Koch 1997) The biological nature of this dormancy is not well understood. However, this feature is central to allowing bacteria to remain in a host for years following the initial infection, a state referred to as latent tuberculosis. (Colston 1999) This makes bacterial sterilization of a host very difficult, particularly since many of the antituberculosis drugs are active only on dividing cells. (Smith 1994) Drug treatment will usually kill most of the mycobacterial population within several weeks; however, the mycobacterial persisters will evade the drug by entering into a dormant state and remain in the host. These remaining bacilli can later lead to reactivation of the infection and post-primary tuberculosis. (Colston 1999)

Our model predicts that one strain is cleared at a significantly slower rate. This reinforces the idea that the mycobacterial population is not of equal drug susceptibility, and that long-term treatment is required in order to clear the persisting mycobacteria.

Our model predicts, based on the observations during the first month of drug treatment, how long it will take to clear the infection. We also predict what proportion of the initial bacterial population is cleared at a slower rate. Unfortunately, as we are not able to distinguish mathematically between a strain that has a decreased death rate and one with an increased birth rate, we cannot precisely estimate the birth and death rate of these strains. Instead, we offer several plausible models that give a range of values for these rates. A more precise estimation of the *in vivo* mycobacterial birth and death rates requires further experimentation. S

Figure 15-1. Model parameters.

Model 1: $N_t = N_1 e^{r_1 t} + N_2 e^{r_2 t}$

Parameter	Explanation	Estimation	95% Confidence Interval
N_t	population size at time= t	----	----
N_1 (A3)	Proportion strain 1at t=0	**0.09**	**[0.044, 0.18]**
N_2 (5E7-N1)	Proportion strain 2 at t=0	0.91	[0.82, 0.95]
r^*_1 (A1)	growth rate strain 1	**-0.166/day**	**[-0.2,-0.1]**
r^*_2 (A2)	growth rate strain 2	**-1.4 / day**	**[-3.2, -0.7]**

Model 2: $N_t = N_1 e^{(\mu - D_1)t} + N_2 e^{(\mu - D_2)t}$

Parameter	Explanation	Estimation	95% Confidence Interval
N_t	Population size at time= t	----	----
N_1 / N_0 *(A1)*	Proportion strain 1at t=0	**0.911**	**(0.79, 0.95)**
N_2 / N_0 *(5E7-N1)*	Proportion strain 2 at t=0	0.099	(0.05, 0.21)
μ *(A4)*	Birth rate	**0.91**	**(0.86, 0.96)**
D_1 *(A3)*	Death rate strain 1	**1.09**	**(1.02, 1.12)**
D_2 *(A2)*	Death rate strain 2	**1.99**	**(1.13, +)**
μ / D_1 *(A3)*		**0.84**	**(0.81, 0.89)**
μ / D_2 *(A2)*		**0.39**	**(0 , 0.77)**

Model 3: $N_t = N_1 e^{(\mu_1 - D)t} + N_2 e^{(\mu_2 - D)t}$

Parameter	Explanation	Estimation	95% Confidence Interval
N_t	Population size at time= t	----	----
N_1 / N_0	Size strain 1at t=0	0.05	
N_2 / N_0	Size strain 2 at t=0	0.95	
μ_1	Birth rate strain 1	0.01	
μ_2	Birth rate strain 1	1.24	
D	Death rate	1.41	

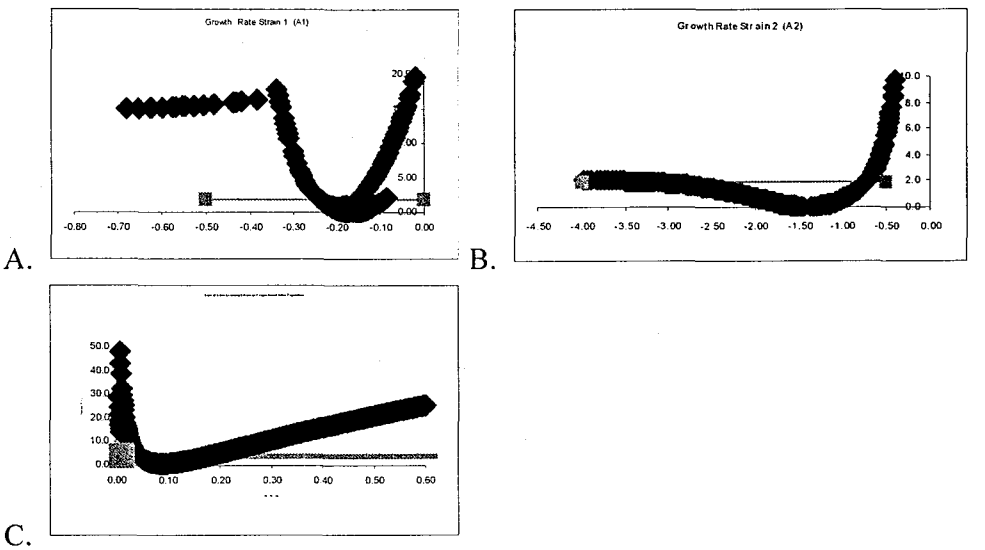
Model 4: $N_t = N_1 e^{\mu_1 (1-d)t} + N_2 e^{\mu_2 (1-d)t}$

Parameter	Explanation	Estimation	95% Confidence Interval
N_t	Population size at time= t	----	----

N_1 / N_0 *(A4)*	Proportion strain 1 at t=0	**0.05**	**[0, 0.15]**
N_2 / N_0 *(5e7 – N2)*	Proportion strain 2 at t=0	0.95	[0.85, 1]
μ_1 *(A2)*	Birth rate strain 1	**0.13 / day**	**[0.05, 0.21]**
μ_2 *(A1)*	Birth rate strain 2	**1.2 / day**	**[0.81, 1.64]**
d *(A3)*	Death rate constant	**1.99 μ**	**[1.58, 2.44]**

$D_1 = (0.079, 0.51)$
$D_2 = (1.27, 4.00)$
$\mu / D = (0.41, 0.633)$

Model 1.



A. B.

C.

Figure 15-2. Model 1. 95% Confidence intervals for Model 1 parameters.

Growth rates of strain 1 (r_1 , A) and strain 2 (r_2 , B), and the proportion of initial population which is cleared at a slower rate (N_1/N_0 , C).

Predicted vs. observed values for model 1.

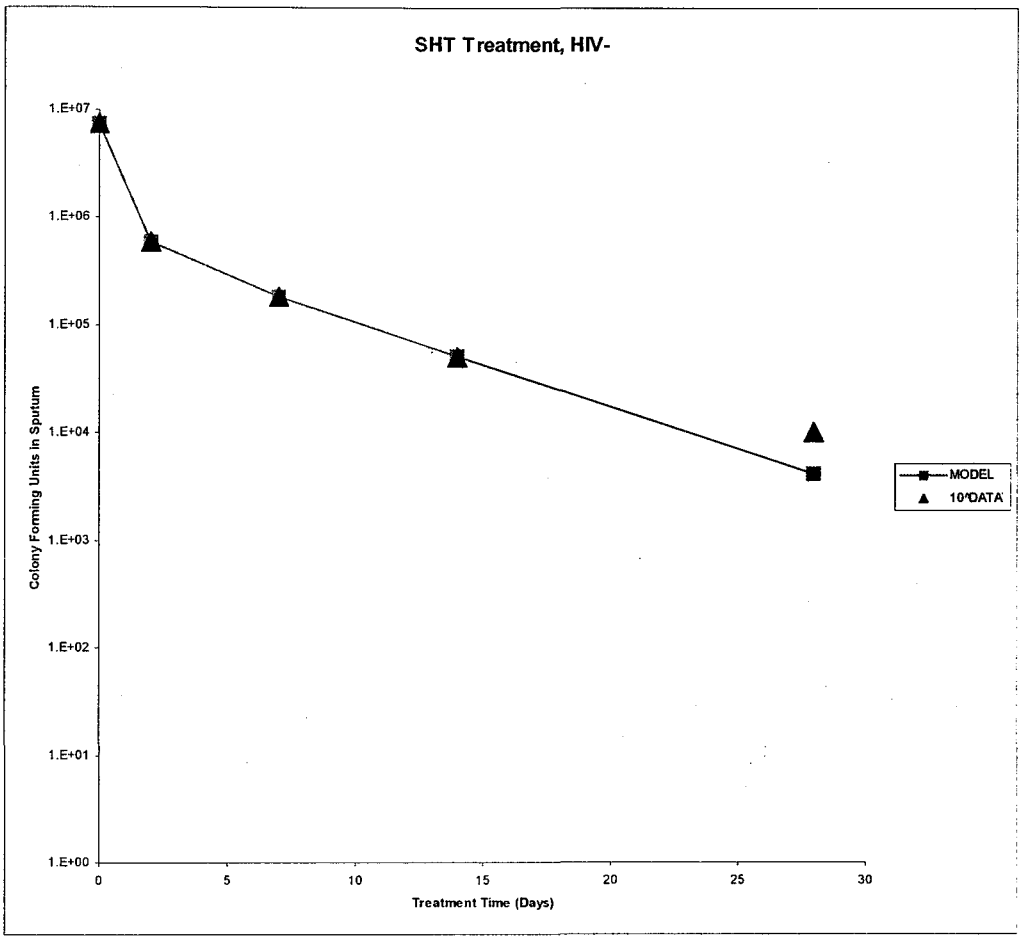
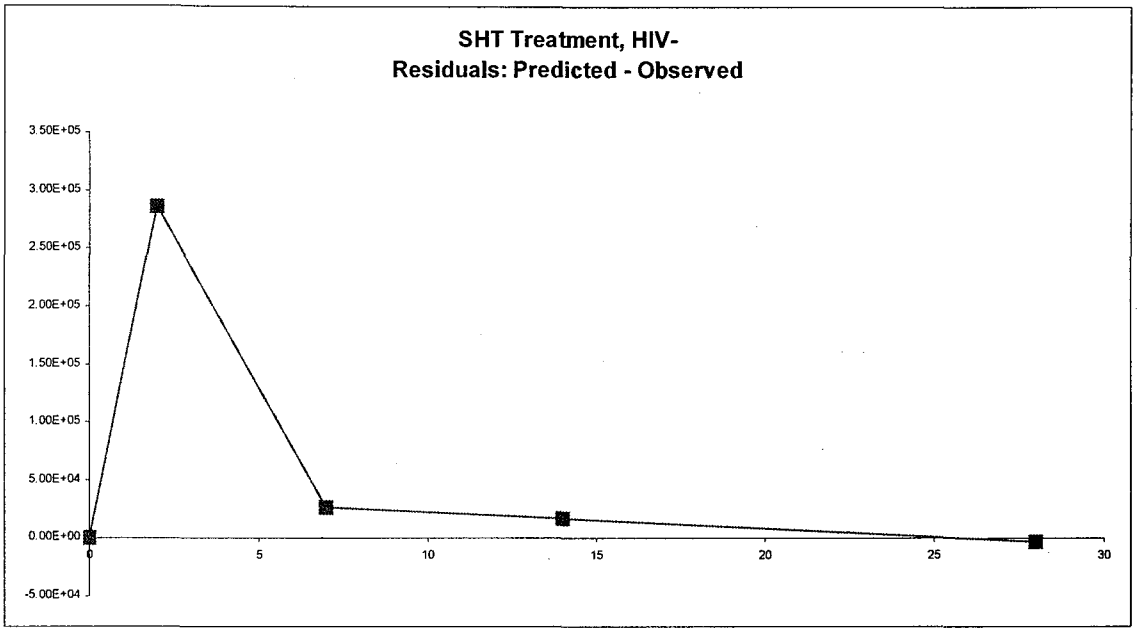


Figure 15-3. Mean data and model 1 predicted values. The mean values of patients in the SHT, HIV- treatment group at each time point are shown above (solid triangles) against the values predicted by model 1 (solid line). The parameters used in model 1 are the best fit parameters from the estimation, as shown in Figure 15-2.

Residuals for Model 1.

A.



B.

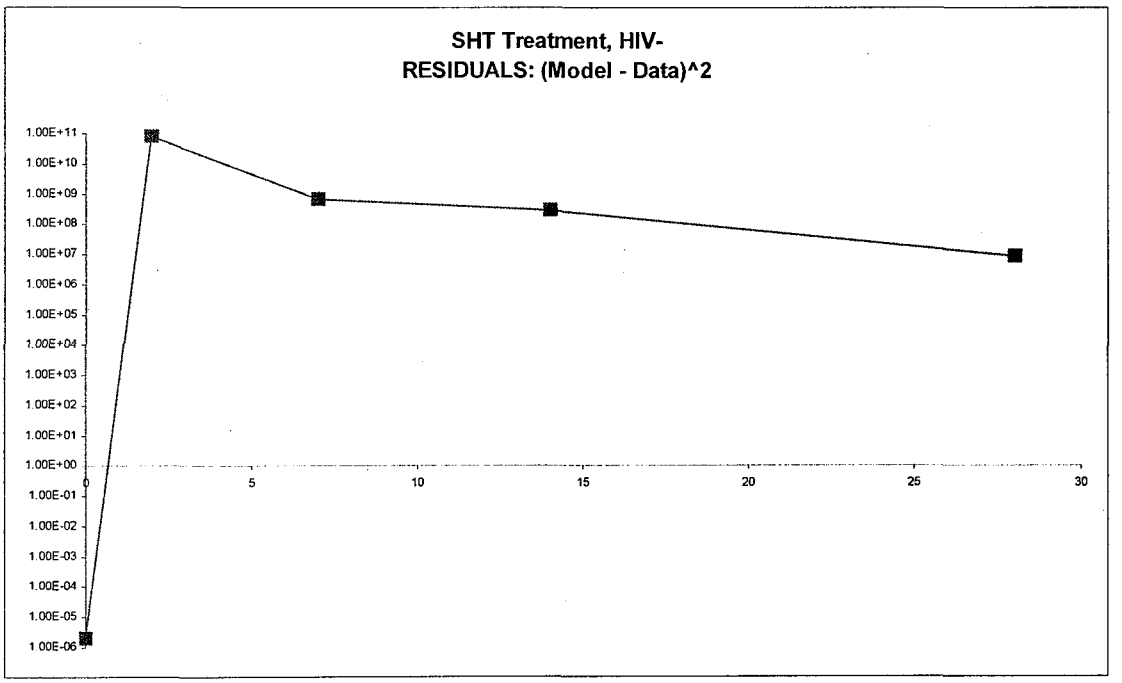


Figure 15-4. Residuals for model 1. The model 1 residuals (predicted – observed), are shown in (A) and the square of these residuals in (B).

Model 2

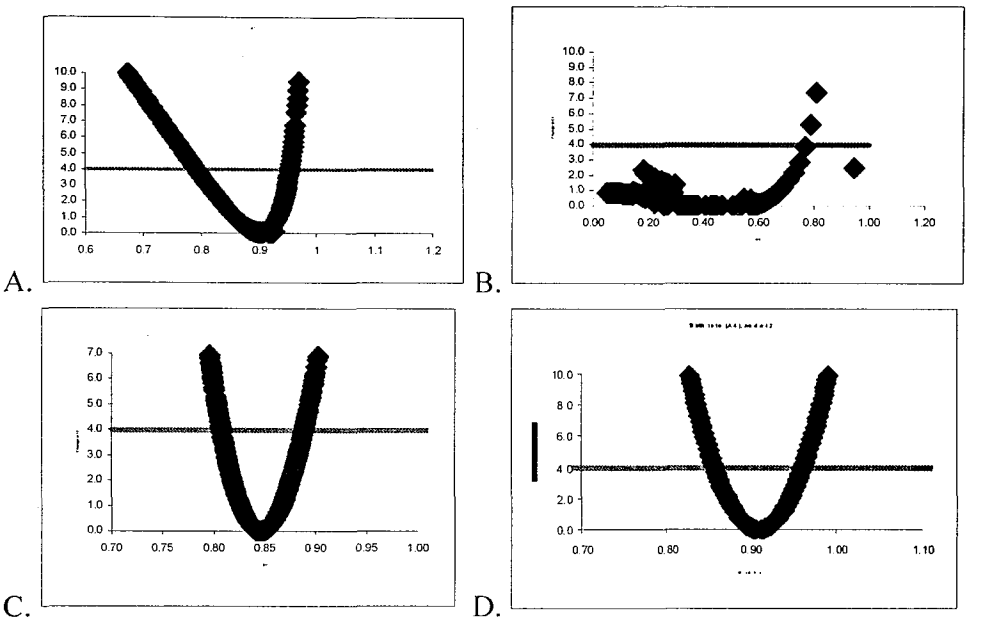


Figure 15-5. Model 2. Confidence intervals for model 2 parameters.

Proportion of initial population which is slow growing, N1/No (A), birth rate μ (D), and the ratio of birth rate to death rate of strain 1 (μ/D1, B) and strain 2 (μ/.D2, C).

Model 4.

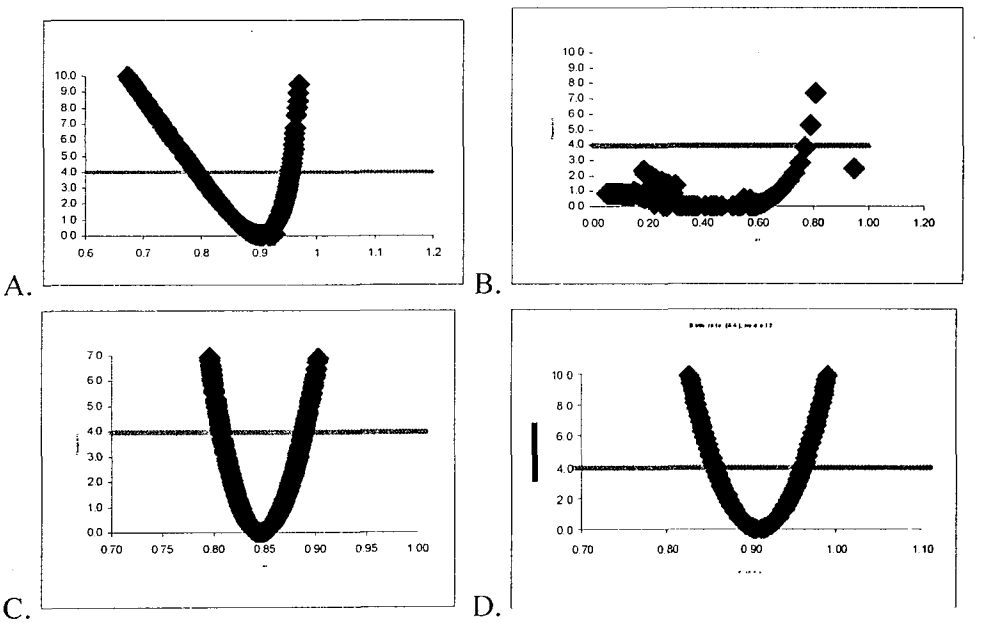


Figure 15-6. Model 4. Confidence intervals for model 4 parameters.

Proportion of initial population which is slow growing, N1/No (A), birth rate μ (D), and the ratio of birth rate to death rate of strain 1 (μ/D1, B) and strain 2 (μ/.D2, C).

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