DEPARTMENT OF COMMUNITY HEALTH
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THE PROGNOSIS OF TUBERCULOSIS IN ADULTS INFECTED WITH HUMAN IMMUNODEFICIENCY VIRUS (HIV-1)

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(M.Sc. STATISTICS)

SUBMITTED FOR THE FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF MEDICINE IN COMMUNITY HEALTH

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Declaration

I, Motasim Badri, hereby declare that the work on which this dissertation is based is my original work (except were acknowledgements indicate otherwise), and that neither the whole work nor any part of it has been, or is being, submitted for another degree in this or any other university.

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ACKNOWLEDGEMENT

Many persons contributed immensely to the study, and are gratefully acknowledged.

I am indebted to Professor Gary Maartens and Dr. Rodney Ehrlich for their dedicated supervision, guidance, willingness and enthusiasm. The work in this thesis is a product of their constructive criticism and insightful suggestions. Their expertise was invaluable in planning and guiding the various phases of this study. I am grateful for their generous contribution of time and resources for accomplishing this study.

Dr. Robin Wood, Head of the HIV clinic at Somerset Hospital, is thanked for making available the database on which this thesis is based, and for his kind advice.

Todd Pulerwitz, Harvard School of Medicine, was a primary collaborator in data collection and validation. He is thanked for his unrelenting efforts in this regard.

The assistance of Dr. Frank Post, Department of Immunology, UCT, in confirming TB diagnosis is appreciated. Professor Jonny Myers, head of the

Department of Community Health provided constructive criticism at the early stages of the study.

Esme Jordaan, the Medical Research Council of South Africa (MRC), and Dr. Jacqui Sommerville, Information Technology Services, Faculty of Engineering, UCT, are thanked for their helpful suggestions and assistance with data analysis techniques.

I am grateful to the World Health Organisation (WHO) for their generous scholarship. Dr Agu Vincent, Administrative officer at the WHO office, Pretoria, is thanked for his kind encouragement and support.

Finally, I am grateful to all my teachers and classmates in the MPhil 1997-98 programme at UCT for their constructive criticism in the early stages of this study, and especially to Mrs. Francesca Little, Dept of Statistics, UCT, for the competent and efficient training in Biostatistics that she provided.

Motasim Hassan Yousof Badri

Cape Town, September 1999

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ABSTRACT

Objectives:

The primary objective of this study was to assess whether active tuberculosis (TB) accelerates the course of HIV-1 infection by measuring progression to AIDS and mortality in HIV-infected patients. Secondary objectives were to evaluate whether TB should be considered an AIDS-defining illness in an area with a high prevalence of TB, and to assess the risk factors for developing TB in HIV-1 infected patients.

Setting:

New Somerset and Groote Schuur Hospital adult HIV clinics, Department of Medicine, Faculty of Health Sciences, University of Cape Town.

Design:

Prospective patient cohort study with five years of follow-up.

Sample:

Adult HIV-infected patients presenting to the two HIV clinics between 1992 and 1996.

Methods:

The TB case definition was a positive culture or a compatible clinical picture combined with a positive smear or a histologic diagnosis. TB patients were treated with 6-month short course regimens.

The Kaplan-Meier method was used to estimate the overall survival times of tuberculosis and non-tuberculosis patients. The generalized log rank test was used to compare the survival curves of these two groups.

The Cox proportional hazards regression method was used to determine the risk of death associated with tuberculosis while adjusting for potential confounding variables (i.e. age, CD4+ count, history of an AIDS-defining illness, use of co-trimoxazole prophylaxis, and antiretroviral therapy, etc.).

The Kaplan-Meier method was used to evaluate the prognosis of HIV-infected patients with TB *vs* the prognosis of HIV-infected with other HIV (or AIDS, as defined by the WHO or the CDC staging systems) related diseases at baseline. The generalized log rank test was used to compare the survival curves of the TB group *vs* the other groups.

Predictors of active TB in HIV-infected patients were assessed using univariate and multivariate logistic regression models.

Results:

Between 1992 and 1996, 609 HIV-infected patients were recruited; of whom 158 (25.9%) were diagnosed with active TB, and 451 (74.1%) were free of active TB by the end of the study. In the TB group, 113 (71.5%) patients presented with active TB at baseline (i.e. prevalent cases) and 45 (28.5%) were free of TB at inclusion but developed active disease during follow-up (i.e. incident cases). Follow-up in the two groups was similar (mean = 11.5 and 14.6 months in cases and non-cases

respectively; $p = 0.7$). The proportion of patients lost to follow-up in the two groups was not significantly different [59 (37%) cases *vs* 153 (34%) non-cases; $p = 0.5$]. In the general cohort 38 patients were suspected for TB but did not fulfill TB case definition and were thus excluded from the study. Of those with TB, 90 (57%) presented with pulmonary TB without evidence of extrapulmonary involvment, 37 (23.4%) presented with pulmonary and documented evidence of extrapulmonary involvement and 31 (19.6%) presented with extrapulmonary alone. Active tuberculosis was confirmed in 69 (43.7%), 80 (50.6%) and 9 (5.7%) patients by culture, smear, and histologic diagnosis respectively.

The risk of developing active TB was significantly increased by: WHO stages 3 or 4, CD4 count <200 cells/μL, presence of one or more AIDS-defining illness at baseline, residence in lower socioeconomic areas and member of a high prevalence TB population.

The proportion of patients presenting at baseline with advanced immunosuppression (i.e.CD4+ T-lymphocyte count < 200 cells/μL) was higher in TB cases [71 (45%)] than in non-cases [145 (33%)] [$p < 0.001$]. TB cases had significantly lower median CD4+ lymphocyte counts compared to non-cases; [130 *vs* 295 cells/μL respectively; $p < 0.01$]. The proportion of TB cases presenting with an AIDS-defining illness at baseline was also higher than that of non-cases [17 (10.8%) TB cases *vs* 24 (5.3%) non-cases; $p = 0.012$].

During the course of follow-up, TB cases developed new AIDS-defining illness more frequently than non-cases; (*risk ratio* RR = 4.10, 95% confidence interval = 2.65-

6.07). The median time of progression to AIDS was significantly different between the two groups. TB cases progressed to AIDS faster than non-cases (median time to AIDS = 6 and 14.5 months respectively; $p < 0.05$). Even after adjusting for confounding, TB was an independent predictor of AIDS (RR= 1.61; 95% confidence interval = 1.08-2.41; p = 0.02).

There was significant depression of CD4+ T-lymphocyte count at the time of diagnosis of active TB. Median CD4+ T-lymphocyte counts *before*, *at* and *after* diagnosis of active TB were significantly different ($p = 0.035$).

TB cases had a reduced survival relative to non-cases; (23.9 and 47.7 months respectively, $p < 0.001$). Active TB was an independent predictor of mortality even after adjusting for confounding; *risk ratio* (RR) = 2.37 (95% confidence interval = 1.46, 3.86). However, after controlling for the transient reduction in the CD4+ T-lymphocyte count induced by active tuberculosis the risk ratio was reduced to 1.70 (95% confidence interval = 1.05-2.77). Other factors associated with increased mortality were CD4+T-lymphocyte count ≤ 200 cells/μ, and history with opportunistic infections at inclusion. Only co-trimoxazole prophylaxis was associated with reduced mortality.

The prognosis of TB patients was different from the prognosis of patients with AIDS ($P < 0.0001$). Survival of TB cases was comparable to survival of patients with oral hairy leukoplakia or oral candidiasis (WHO stage 3, CDC stage B).

Of the 158 HIV-infected patients with TB, 71(44.9%) patients used co-trimoxazole prophylaxis and 87 (55.1%) did not. Co-trimoxazole prophylaxis reduced the incidence of AIDS-defining illnesses which could be prevented by co-trimoxazole (risk ratio = 0.23; 95 % confidence interval = 0.06-0.84; p = 0.017). Co-trimoxazole prophylaxis was protective against mortality even after adjusting for confounding (risk ratio = 0.53; 95% confidence interval = 0.30-0.94; p = 0.03). The better outcome in patients given co-trimoxazole prophylaxis was achieved despite their lower CD4+ lymphocyte count at baseline than patients not given the drug.

Conclusion :

Active TB accelerates HIV-1 disease progression as indicated by the increased incidence of AIDS-defining illness and the increased mortality following TB diagnosis observed in patients of this cohort. All HIV-infected patients should thus be routinely screened for TB and effective prophylaxis should be considered to prevent TB.

According to the findings of this study, HIV-infected patients presenting with WHO stages 3 or 4, a CD4 count < 200 cells/μL, presence of one or more AIDS-defining illness, residence in lower socioeconomic area or who belong to communities with higher TB prevalence must be considered as candidates for developing active TB. These categories of HIV-infected patients should be specifically targeted for TB prophylaxis.

Co-trimoxazole prophylaxis reduces the frequency of AIDS-defining illness and improves the survival of HIV-infected patients with CD4+ T lymphocyte < 200 or

with TB. Its administration to HIV-infected patients with CD4+ T lymphocyte < 200 or with TB is strongly recommended.

Despite the accelerated course of HIV infection following its diagnosis, however, TB prognosis in this cohort was significantly better than that of the other AIDS-defining illnesses. Thus in endemic areas TB should not be included in the case definition of AIDS.

1. INTRODUCTION

This thesis sets out to examine, by way of a prospective cohort study, the prognostic effect of tuberculosis in adult patients with Human Immunodeficiency Virus (HIV-1) infection in Cape Town.

The foundations of this thesis have been laid over the past 14 years, during the course of which a total of more than 8,868 visits were made by a cohort of more than 1288 patients who were seen or treated in the two HIV clinics at the New Somerset (NSH) and Groote Schuur (GSH) hospitals, two tertiary hospitals affiliated to the Faculty of Health Sciences, University of Cape Town.

When the first AIDS cases were identified in Cape Town in 1984, a multidisciplinary HIV clinic was developed and supervised by the late Dr. Frank Spracklen at the New Somerset Hospital; followed by a second clinic at the Groote Schuur Hospital. These clinics drew patients from across the sociodemographic spectrum of the Western Cape province [1].

Cape Town is at the intersection of two epidemics: tuberculosis prevalence and incidence rates are among the highest in the world, and HIV infection is rapidly spreading in the population and reaching exponential growth. A surge of tuberculosis

can be anticipated, especially in the Black* population, as HIV becomes more prevalent, and more HIV infected persons develop significant immune compromise. Data from New Somerset Hospital, one of the study sites, revealed a large increase in tuberculosis admissions between 1990 and 1995. This increase was entirely accounted for by HIV positive patients, whereas the number of HIV negative patients with tuberculosis remained unchanged [2]. The overall HIV prevalence rate among patients admitted to Somerset Hospital with tuberculosis and prior unknown HIV status was 39 % in 1994-1996: 37 % in Coloured and 53 % in Black patients [2].

Worldwide, many investigators have assessed the survival times of patients with HIV/AIDS and the factors which influence their prognosis. A clear understanding of the prognosis of patients with HIV/AIDS, and the factors that influence survival, allows an insight into the pathophysiology of this disease, evaluation of the impact of new treatments and the development of interventions which may improve survival of current or future patients.

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* Under apartheid, South Africans were classified into four "race" groups: Asian, Black, Coloured, and White. These groups signify, to varying degrees, differences in socioeconomic status, cultural practices and urbanisation.

Factors associated with survival in patients with HIV/AIDS can be summarized, according to the available evidence gained from a large number of studies done so far in different settings, as:

A : Strong evidence for association with decreased survival :

1. Greater age [3-11];
2. Initial AIDS-defining illness [3-14];
3. Increased number of subsequent AIDS-defining illnesses[3,5,14];
4. Lower CD4+ lymphocyte count at diagnosis [3,6-7,13-15];
5. Loss of CD4+ cells after diagnosis [17-18];

B: Inadequate evidence for association with decreased survival :

1. Pregnancy [19];
2. Poorer socioeconomic circumstances [15,20-24];
3. Irregular follow-up care [25];
4. Co-infection with other viruses [26-31];
5. Increased number of sexual partners following diagnosis [32-34];
6. Depression [35];
7. Smoking [36];
8. Recreational drug use [33];
9. Greater alcohol intake [33];

C: Evidence against an association

1. Gender [3,4,11,36];
2. Ethnic origin (within developed countries) [3,4,7,9,12];

Persons with HIV infection are at risk of developing numerous AIDS-defining illnesses. A large percentage of individuals with HIV infection suffer AIDS-defining

illness before death. Of importance in the wide spectrum of these diseases is tuberculosis, which has been reported as the most common serious HIV-related complication worldwide.

The prognosis of HIV-infected patients with tuberculosis has been difficult to assess. Firstly, tuberculosis occurs commonly in non-immunocompromised persons and therefore a diagnosis of tuberculosis does not necessarily indicate immunodeficiency. Secondly, active tuberculosis, if not treated, can decrease the number of circulating CD4+ T-lymphocytes, thereby interfering with one of the best predictors of survival in HIV infection. If active tuberculosis is successfully treated, however, a significant increase in the CD4+ T-lymphocyte may be achieved [18]; and this may have a positive impact on the prognosis of HIV-infected patients.

The clinical importance of tuberculosis in the spectrum of AIDS-related disease is reflected in the revised case definition of AIDS of the Centers for Disease Control and prevention (CDC) (Appendix I). This definition includes pulmonary tuberculosis as an AIDS-qualifying diagnosis in HIV-infected patients. This is due to the fact that in the USA and the other developed countries TB confers a prognosis similar to the other AIDS-defining conditions [37].

Given the exponential growth of HIV infection in Cape Town and the persistently high notification rate of tuberculosis [Table 1], evidence of the association between active tuberculosis and HIV-infection may be easier to study in such a setting. The Cape Town population includes a wide spectrum of people with different characteristics. Furthermore, the Western Cape has one of the best-functioning TB

notification systems in Africa. This makes possible thorough, comparative investigation into the role which tuberculosis may have on the progression of HIV infection.

As discussed later, the findings of the studies done so far regarding the prognosis of HIV-related tuberculosis are inconsistent and, in some cases, contradictory. There is a need for further studies in a high tuberculosis prevalence setting where TB occurs in HIV-infected patients at different levels of immune suppression, and where childhood infection may mean better TB immunity. Such studies may shed light on the mechanisms of interaction between these two intracellular pathogens, and lead to novel strategies to improve survival of HIV-infected patients with tuberculosis.

Table 1.1
Notification of tuberculosis in the Western Cape, per 1000 of population: 1989-96.

Population Group	1989/90	1990/91	1991/92	1992/93	1993/94	1994/95	1995/96
White	0.2	0.17	0.20	0.23	0.19	0.17	0.20
Coloured	4.19	4.33	4.22	4.13	3.82	4.02	3.92
Asian	0.55	0.47	0.22	0.06	0.22	0.23	0.15
Black	8.89	9.02	8.32	8.67	7.53	7.93	8.11
Total	3.94	4.05	3.89	3.91	3.55	3.72	3.74

(Ministry of Health, Cape Town).

1-2 AIM:

The aim of this study was to measure the survival time, incidence of AIDS-defining illness and mortality rate, and their associations with various risk factors, among HIV patients co-infected with active tuberculosis and HIV in two adult HIV clinics affiliated to the University of Cape Town.

1-3 OBJECTIVES:

A. Main objectives :

1. To assess whether HIV-infected clinic patients with active tuberculosis have an overall shorter survival than HIV-infected patients without active tuberculosis.
2. To determine whether HIV-infected patients with active tuberculosis develop other AIDS-defining illnesses at a greater rate than HIV-infected patients without active tuberculosis.
3. To assess whether site of tuberculosis (pulmonary / extrapulmonary or both) influences or predicts the survival of HIV-patients with active tuberculosis.

B. Primary objectives:

1. To measure the risk of developing active tuberculous disease in HIV-infected clinic patients.
2. To ascertain predictors of active tuberculous disease in HIV-infected clinic patients.
3. To ascertain whether active tuberculosis, at the time of diagnosis, induces a loss in the circulating CD4+ T lymphocytes in HIV-infected clinic patients.
4. To describe the sociodemographic profile of HIV-infected patients with active tuberculosis compared with HIV-infected patients without active tuberculosis.

1-4 STRUCTURE OF THESIS:

This thesis is divided into five parts. Chapter1 (above) gives an introduction to the setting in which this study was done along with a brief history of the two HIV clinics from which the study cohort was drawn. Chapter1 also includes an introduction to the problem and a summary of the factors so far known to be associated with survival in patients with HIV/AIDS. The aim and objectives of the thesis are set out.

Chapter 2 is a review of the literature, particularly of studies of relevance to the objectives of this study.

Chapter 3 describes the methods, measurements and statistical techniques used in data analysis. This chapter also specifies the confounding and effect modifier variables which might influence the association between active tuberculosis and HIV infection.

Chapter 4 sets out the results. The baseline characteristics of the patients are described, with a brief discussion of the differences observed between the two comparison groups (i.e. active TB cases and "controls" without TB). Results of the survival analysis are presented in tabular and graphical format.

In chapter 5, the findings and limitations of this study are discussed and compared with the findings of other studies done in different countries. Recommendations for health care policy and further research are included at the end of this chapter.

In chapter 5, the findings and limitations of this study are discussed and compared with the findings of other studies done in different countries. Recommendations for health care policy and further research are included at the end of this chapter.

2. LITERATURE REVIEW

In the last decades of this century AIDS has emerged as a major cause of morbidity and mortality in many countries. It is estimated that there are about 22 million persons in the world living with HIV/AIDS. 14 million of them are thought to be in sub-Saharan African countries, representing about 63% of the world's total cases. In urban centers of central Africa 5 to 20% of the sexually active population is infected with HIV [38].

On the other hand, tuberculin surveys have shown that it is likely that one third of the world's population, i.e. 1700 million people, has been infected with the tubercle bacillus. From this infected pool, 8-10 million people develop active tuberculosis annually resulting in 2.9 million deaths. As the disease is a chronic one and treatment is often delayed or inadequate, there is more than this number of patients with infectious disease. As, on average, each such person infects 20 people annually, about 100 million people are added to the infected pool each year [39]. The number of new tuberculosis cases occurring each year was predicted to increase from 7.5 million (143 cases per 100,000) in 1990 to 8.8 million (152 per 100,000) in 1995 and 10.2 million (163 per 100,000) in the year 2000. 2.5 million persons were estimated to have died of tuberculosis in 1990, 3 million in 1995, with 3.5 million deaths from tuberculosis

predicted to occur in 2000 (Table 2.2). Tuberculosis is responsible for 1 in every 4 preventable adult deaths, despite being one of the most cost-effective of adult diseases to treat.

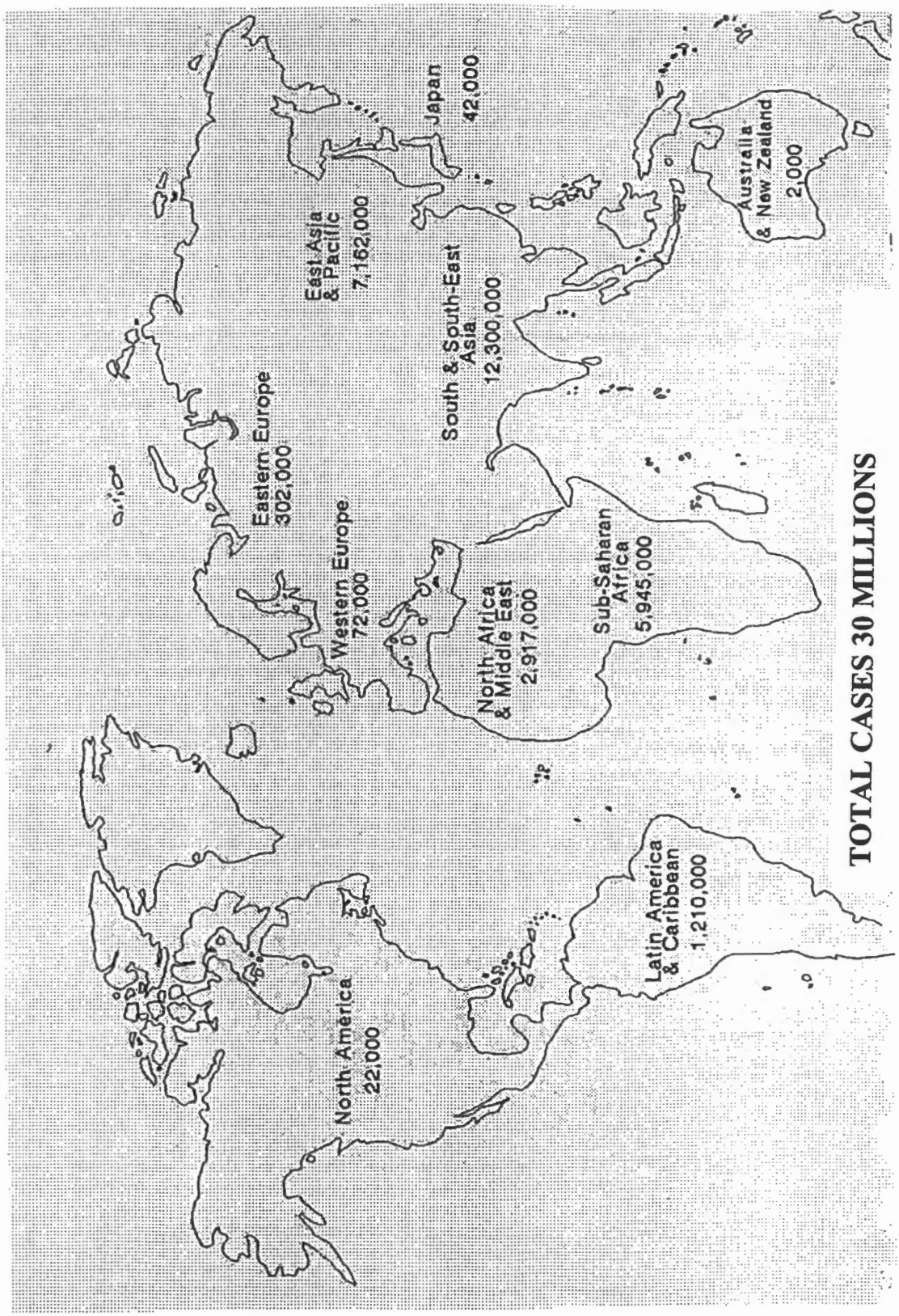
2-1 HIV-RELATED TUBERCULOSIS EPIDEMIOLOGY

Since the mid-1980s it has been evident that the HIV/AIDS pandemic is having a devastating effect on the prevalence of tuberculosis. The reason for concern is that persons infected with both the tubercle bacillus and HIV have a much higher chance of developing active tuberculosis than HIV-negative persons. It is estimated that only 5 percent of persons who have successfully overcome the primary infection develop active post-primary tuberculosis during the remainder of their lives, which may extend over several decades. By contrast, about 50 per cent of HIV-positive persons infected with the tubercle bacillus develop the disease over a life span considerably shortened by the other effects of HIV. Thus some 8 per cent of HIV-positive persons infected by the tubercle bacillus develop active tuberculosis annually, a 20-fold higher rate than in the HIV-negative groups [40].

Dolin *et al.* have noted that 95 per cent of cases of tuberculosis in the dually infected population are the result of the HIV infection while the remaining 5 per cent would have occurred if the patients had not been infected with HIV [41]. From data on the prevalence of both infections, it was calculated that, in mid-1994, about 5.6 million people worldwide were dually infected [42], and that HIV caused an additional 450,000 cases of tuberculosis in 1994, with over 300,000 of these in the sub-Saharan Africa. In that year, at least 5 per cent of all cases of tuberculosis worldwide and over

20 per cent of cases in Africa were HIV-related. It has been estimated that by the year 2000 there will be 1,410,000 cases of HIV-related tuberculosis worldwide (604,000 in Africa); accounting for about 14 per cent of all cases of tuberculosis (Table 2.1 and Table 2.2).

In summary, the HIV pandemic will worsen the tuberculosis situation in developing countries in three ways: (1) by reactivation of latent tuberculosis infection among dually infected persons; (2) by new infection with tubercle bacilli and rapid progression to active disease in HIV-infected persons; and (3) by increasing the number of cases in the general population whose infection and disease will result from transmission from HIV-positive individuals developing tuberculosis by either reactivation or recent infection [42].


TOTAL CASES 30 MILLIONS
North America
22,000
Western Europe
72,000
Eastern Europe
302,000
East Asia
& Pacific
7,162,000
Japan
42,000
South & South-East
Asia
12,300,000
North Africa
& Middle East
2,917,000
Sub-Saharan
Africa
5,945,000
Latin America
& Caribbean
1,210,000
Australia
& New Zealand
2,000

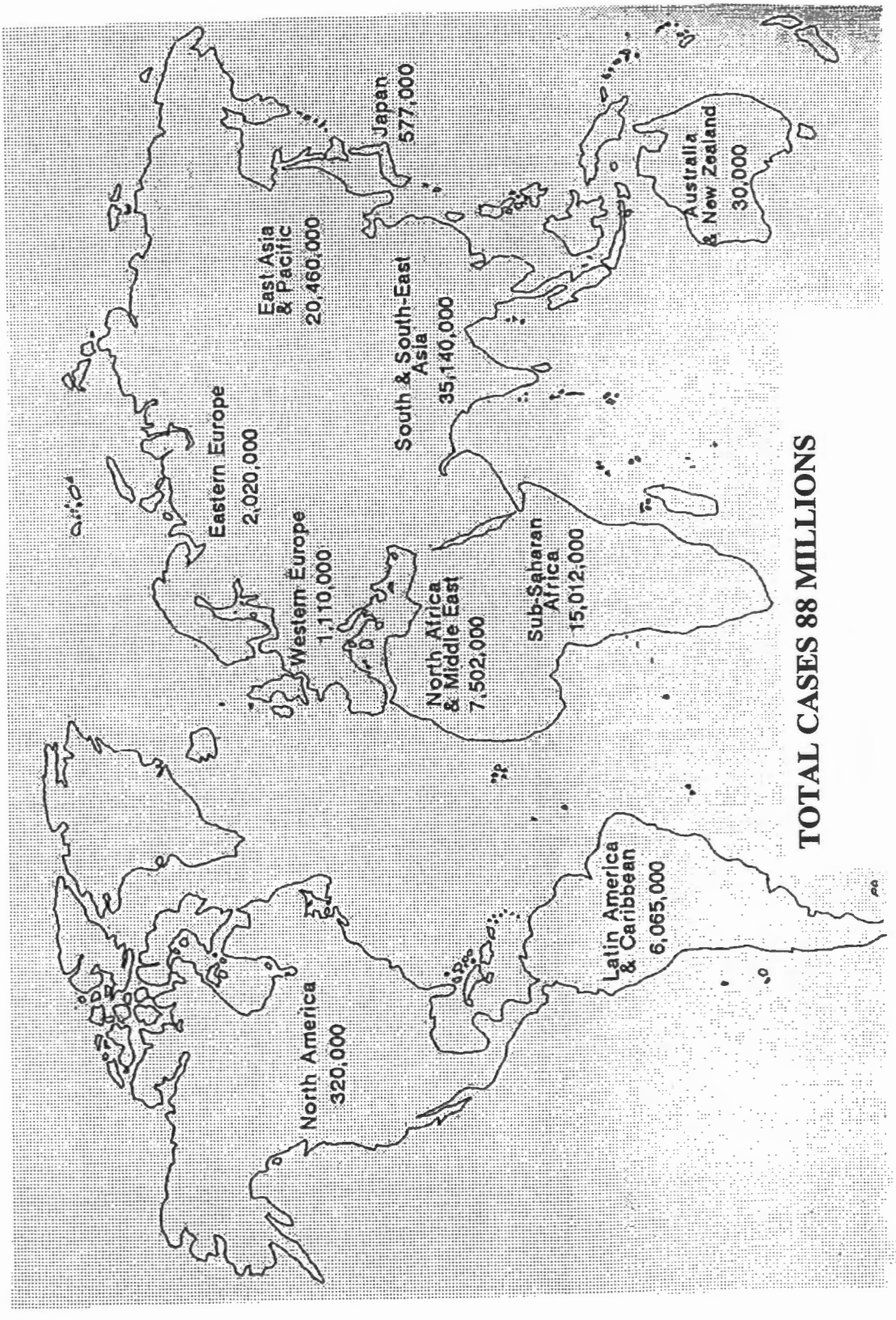

TOTAL CASES 88 MILLIONS
North America
320,000
Latin America
& Caribbean
6,065,000
North Africa
& Middle East
7,502,000
Sub-Saharan
Africa
15,012,000
Western Europe
1,110,000
Eastern Europe
2,020,000
East Asia
& Pacific
20,460,000
Japan
577,000
South & South-East
Asia
35,140,000
Australia
& New Zealand
30,000

Table2.1
Estimated tuberculosis incidence and HIV-attributable tuberculosis in 1990,1995 and 2000, by world regions

Region	1990			1995			2000		
	Total TB Cases	Rate[a]	HIV-Attributed TB cases	Total TB cases	Rate[a]	HIV-Attributed TB cases	Total TB Cases	Rate[a]	HIV-Attributed TB cases
South-East Asia	3,106,000	237	66,000	3,499,000	241	251,000	3,952,000	247	
Western Pacific[b]	1,839,000	136	19,000	2,045,000	140	31,000	2,255,000	144	68,000
Africa	992,000	191	194,000	1,467,000	242	380,000	2,079,000	293	604,000
Eastern Mediterranean	641,000	165	9,000	745,000	168	16,000	870,000	168	38,000
Americas[c]	569,000	127	20,000	606,000	123	45,000	645,000	120	97,000
Eastern Europe[d]	194,000	47	1000	202,000	47	2,000	210,000	48	6,000
Industrialized countries[e]	196,000	23	6,000	204,000	23	13,000	211,000	24	26,000
Total	7,537,000	143	315,000	8,768,000	152	738,000	10,222,000	163	1,410,000
% Cases attributble to HIV			(4.2%)			(8.4%)			(13.8%)
Increase since 1990				16.3%			35.6%		

(P.J. Dolin 1994)

[a] Crude incidence rate per 100,000 population. [b] Includes all countries of the Western Pacific Region of WHO, except Japan, Australia and New Zealand [c] Includes all countries of the American Region of WHO ,except USA and Canada. [d] Eastern European countries , and independent states of former USSR. [e] Western European countries , USA, Canda, Japan, Australia and New Zealand.

Table 2.2
Estimated world tuberculosis mortality and HIV-attributed tuberculosis mortality 1990-2000, by region

Region	Deaths in 1990		Deaths in 1995		Deaths in 2000	
	Total	Attributed To HIV	Total	Attributed To HIV	Total	Attributed To HIV
South-East Asia	1,087,000	23,000	1,225,000	88,000	1,383,000	200,000
Western Pacific[a]	644,000	7,000	716,000	11,000	789,000	24,000
Africa	393,000	77,000	581,000	150,000	823,000	239,000
Eastern Mediter-ranean	249,000	4,000	290,000	6,000	338,000	15,000
Americas[b]	114,000	4,000	121,000	9,000	129,000	19,000
Eastern[c] Europe	29,000	< 200	30,000	< 600	32,000	< 900
Industriali-zed countries[d]	14,000	< 500	14,000	1,000	15,000	2,000
All regions	2,530,000	116,000	2,977,000	266,000	3,509,000	500,000
% deaths attributable to HIV		(4.6 %)		(8.9 %)		(14.2 %)
Increase since 1990			17.7 %		38. 7%	

(Bulletin of WHO 1992:72). [a] Excluding Japan, Australia and New Zealand. [b] Excluding USA and Canada.[c] Eastern Europe and independent states of former USSR.[d] Western Europe, USA, Canada, Japan, Australia and New Zealand.

Although HIV-infected tuberculosis patients respond well to antituberculosis chemotherapy, they have a high short-term mortality [43,44]. In Zaire, the case fatality ratio among HIV-seropositive patients was 31.3% one year after diagnosis compared to 4.4 % for HIV-seronegative patients [43]. Similar results were reported from Central Africa Republic, Zambia and Kenya [44]. Furthermore, data from some programmes in East Africa suggest that mortality among tuberculosis patients has increased significantly since the mid 1980s. A number of reports further suggest that tuberculosis is a common life-threatening complication associated with HIV infection in Africa. In a retrospective cohort study of 249 HIV-seropositive and 310 HIV-

seronegative women of childbearing age in Zaire, 11 % of HIV-seropositive women died during 2 years of follow-up while none of the HIV-seronegative women with proven tuberculosis were dead at the end of the study [45].

Exogenous infection also occurs in HIV-infected individuals and leads to a greatly accelerated progression from infection to disease. In an outbreak of HIV-related tuberculosis due to an unusual multi-drug resistant strain of *M.bovis*, five HIV-positive persons briefly exposed to the source case presented with extensive disease due to the same bacillus 2 to 10 months later [46].

Before the onset of the HIV pandemic the incidence rate of tuberculosis was constant or in slight decline in the developing nations, but owing to population growth, the number of cases of the disease was increasing. Future trends of TB in the world will be determined by the annual tuberculosis infection rate, the prevalence of tuberculosis infection in the age group principally at risk and the HIV prevalence rate. Schultzer *et al.* have calculated that, given an optimistic scenario of a one per cent annual tuberculosis infection rate, a 45 percent prevalence of tuberculosis infection in the at-risk age group and a 2 percent prevalence of HIV infection, there will be a 68 per cent increase in the number of cases of tuberculosis from the 1980 figure by the year 2000. A worse-case scenario with corresponding values of 2, 60 and 20 percent would lead to a 12-fold increase in cases of tuberculosis over the same time period, amounting to an eventual incidence rate over 4,000/100,000 in the at-risk age range, and about 2,000/100,000 of the total population. This implies that one in 50 of the population would develop tuberculosis each year [47].

In industrialized nations, tuberculosis is an uncommon disease. However several developed countries have reported a rising incidence in recent years. Several lines of epidemiological evidence support the hypothesis that the HIV epidemic is an important underlying cause of the resurgence of TB in the developed countries. In the United States the cities with the largest numbers of AIDS cases had the greatest increases in reported cases of tuberculosis. In New York, the city with the largest number of patients with AIDS, the number of reported cases of tuberculosis increased by 68% in 1980 to 1989. In addition the incidence of tuberculosis in patients with AIDS was 500 times the incidence of tuberculosis in the general population. The risk of active tuberculosis among HIV-infected persons with positive tuberculosis skin tests was estimated to be 8% per year [48-49]. In France, 19.9 tuberculosis cases per 100,000 population were reported in 1992, a 6.6% increase compared to the previous year [50]. Table 2.3 shows increase in tuberculosis incidence in some European countries.

Table 2.3
Increase in the incidence of tuberculosis in some industrialized countries.

Country	Increase (%)	Time period
Austria	5	1998-90
Denmark	20	1986-92
Ireland	9	1988-91
Italy	27	1988-92
Netherlands	19	1987-92
Norway	21	1988-92
Spain	28	1990-92
United Kingdom	5	1987-91
USA	20	1985-92

(***World Health Organization*** , **1994**)

2-2 HIV AND TB IN AFRICA

Tuberculosis associated with HIV is reaching epidemic proportions in many African countries [43,51,52]. Here HIV infection is often associated with tuberculosis, leading to a negative effect on the outcomes of both diseases. Studies from the United States and Africa have shown higher mortality rates among patients with tuberculosis and HIV infection than among patients with tuberculosis but without HIV infection, although in the United States this difference has been attributed by some studies to complications of HIV related infections other than tuberculosis [53]. It is not clear if this is also the case in the developing countries [54]. Higher mortality rates from tuberculosis in the developing countries have been attributed to treatment program failures such as drug shortages, non-compliance or higher level of initial resistance, unavailability of short-course rifampicin regimen, any or all of which could lead to the apparent failure of anti-tuberculosis therapy [55]. It was estimated that in 1992 alone, HIV infection was responsible for an additional 150,000-250,000 cases of tuberculosis in Africa, and that the HIV prevalence in African patients with pulmonary tuberculosis was noted to be as high as 70% [44].

Tuberculosis has become by far the commonest opportunistic illness in HIV infected patients in Africa. HIV seroprevalence rates of greater than 40% are common among patients with tuberculosis in many African countries. In Kampala, Uganda, 66% of newly diagnosed tuberculosis patients were HIV-positive [56]; in Zambia, 60% of tuberculosis patients were HIV-infected [51] and in Kenya, 30 % of newly diagnosed tuberculosis patients in Nairobi hospital were HIV-seropositive. Overall, these rates are much higher than those seen in the general population. For example, in Ivory

Coast, 26 % of 2043 tuberculosis patients and 7.2 % of 2127 blood donors were HIV-seropositive [57].

In South Africa, Wood and Post in their follow-up of a cohort of 150 HIV infected patients presenting with pulmonary tuberculosis to a Cape Town HIV clinic between 1989 and 1994, have demonstrated an overall mortality rate of 41/100 person years [58]. Slavik *et al.* carried out a retrospective cross-sectional study in Pretoria to determine the prevalence of HIV positivity in patients with tuberculous lymphadenitis. HIV was often associated with extrapulmonary forms of tuberculosis, lymph nodes being the commonest site affected in their patients. They observed that 54.3 % of the patients who had lymph node biopsies were HIV positive. Their study showed significant increases in HIV positivity among study cases over 9-years period ($p < 0.0001$). They concluded that the escalation in HIV positive cases in 1993 and 1994 was due to the increasing association between HIV infection and tuberculosis being seen in South Africa, and emphasised the importance of screening patients with tuberculosis for HIV infection [59].

In four Ugandan hospitals located outside Kampala, the HIV seroprevalence among 1072 new patients with tuberculosis tested in an unlinked anonymous manner was 43% compared to 16% among 3324 women attending antenatal clinics at the same hospital [56]. Based on these data, it is possible to estimate that between 30% and 40% of incident tuberculosis cases in Africa are attributable to HIV infection.

The burden of the additional HIV-related cases of tuberculosis has been felt in several African countries and health services are being severely stretched. Reported cases of

tuberculosis in four African countries with reliable reporting systems, Tanzania, Burundi, Malawi and Zambia, showed increases of 86%, 140%, 180%, and 154% respectively between 1984 and 1990 [44]. In Zaire, HIV prevalence in patients with confirmed tuberculosis was 17 % in outpatients and 33% in inpatients [43]. This high HIV seroprevalence among inpatients with TB has important implications, as inpatients consume more resources.

The complications of HIV-positive status include a 5-8% annual risk and a 30% or greater lifetime risk of developing tuberculosis for those with a positive tuberculin skin test [55]. Other studies have demonstrated that HIV-infected patients have an incidence rate of active tuberculosis between 3.1 and 7.9 cases per 100 person years [60].

In one African study, a case fatality rate of 77% in patients infected with both HIV and tuberculosis was reported compared to 11 % in patients with tuberculosis without HIV [61]. HIV-positive tuberculosis patients in Africa are almost four times as likely to die of tuberculosis than HIV-negative patients within 13 months of diagnosis, mostly during the first month of treatment [55]. In addition, the long term survival of the cured patients may be shortened because of the synergistic immune suppressive action of the HIV and *M. tuberculosis* [55]. Thus, even if treated, tuberculosis may have a devastating effect on the health and life span of young adults who are essential to the economic stability of their community.

2.3 HIV and TB in South Africa

South Africa is considered to have one of the fastest growing HIV epidemics in the world, with close to 15,000 people infected every month [62]. The countrywide HIV prevalence for South African adults is based on the sampling of women attending ante-natal clinics, which have been considered as suitable sentinel sites for HIV prevalence in South Africa (Fig 2.3). These prevalences were estimated at 14.17% in 1996, and 16.01% in 1997 [63]. This indicates a significant increase compared to 1995 (prevalence = 10.44%) and 1994 (prevalence = 7.57 %). HIV prevalence was shown to be rising in all age groups [Table 2.5]. Within South Africa, large regional variation of HIV positivity has been observed, ranging from 6% in the Western Cape to 26 % in KwaZulu-Natal (Fig 3,4).

Several models have been proposed to predict the extent of the AIDS epidemic in South Africa. The Doyle model predicts that between 18 % and 27 % of the South African population will be HIV-infected by the year 2010 [64]. Data available from country-wide surveys and from sentinel sites have reinforced these predictions. Projections for Soweto, based on the Doyle model, have been within the same range of prevalence rates as those experienced at Baragwanath hospital [65].

FIG 2.3

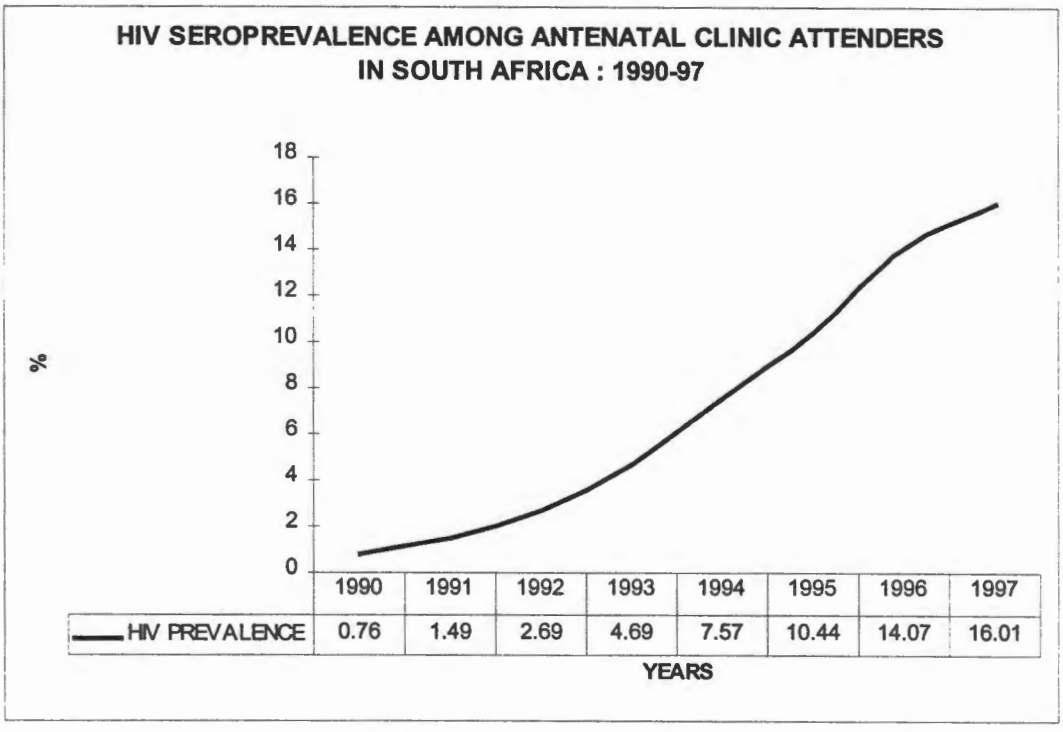
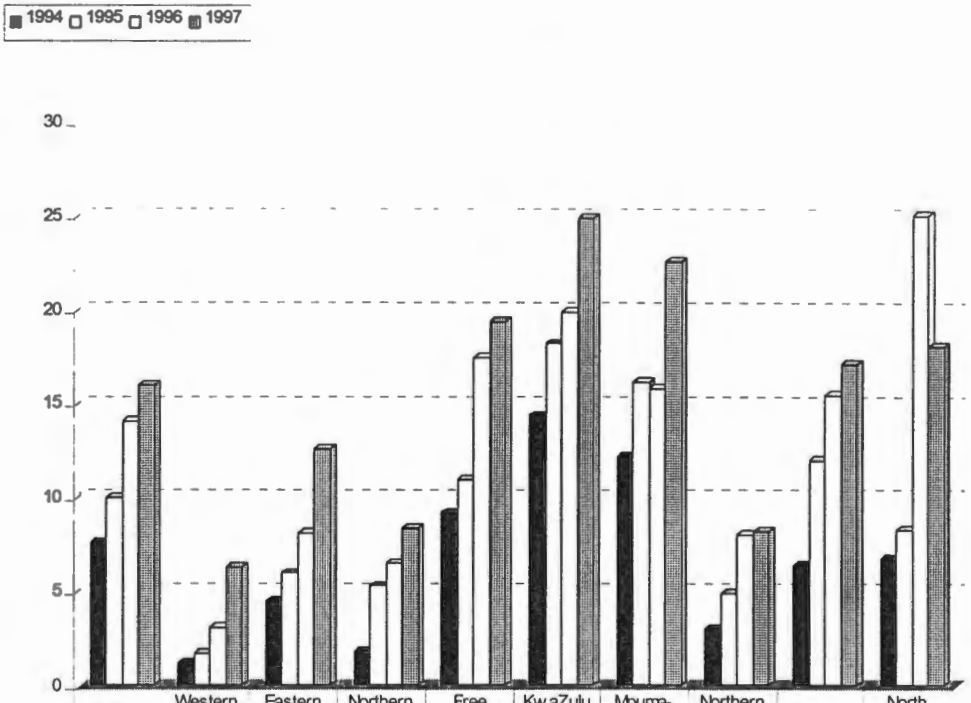


Table 2-4
Prevalence of HIV infection in South Africa by age: 1994-1996

AGE GROUP	1994 % (95% C.I)	1995 % (95% C.I)	1996 % (95% C.I)
<20	6.5 (5.5-7.5)	9.5 (8.2,10.9)	12.8 (11.3,14.2)
20-24	8.9 (8.0,9.9)	13.1 (12.0,14.3)	17.5 (16.3,18.8)
25-29	8.6 (7.3,10.3)	11 (9.8,12,3)	15.2 (13.8,1.5)
30-34	6.4 (5.3,7.5)	8 (6.8,9.3)	12.1 (10.7,13.6)
35-39	3.7 (2.3,5.1)	7.4 (5.5,9.3)	9.7 (7.0,11.5)
40-44	5.3 (3.2,7.4)	4.4 (2.0,6.8)	9.9 (5.8,14.1)
45-49	0.4(0.1,1.2)	7.5 (0.1,19.4)	5.8 (2.3,9.4)

Source : *Epidemiological comments*, October 1996

Fig 2.4
HIV incidence rates In South Africa by province, 1994-1997



	RSA	Western Cape	Eastern Cape	Northern Cape	Free State	KwaZulu Natal	Mpuma-langa	Northern Province	Gauteng	North West
1994	7.6	1.2	4.5	1.8	9.2	14.4	12.2	3	6.4	6.7
1995	10.04	1.7	6	5.3	11	18.2	16.2	4.9	12	8.3
1996	14.1	3.1	8.1	6.5	17.5	19.9	15.8	8	15.5	25.1
1997	16	6.3	12.6	8.4	19.4	24.9	22.6	8.2	17.1	18.1

(Department of Health, annual antenatal survey)

It is estimated that about two thirds of adult South Africans have been exposed to TB. There is thus a large pool of people harbouring dormant TB bacilli. In 1993 the national TB incidence rate was 225/100,000 population, rising to 362/100,000 by 1996 (Table 2.5). More than the quarter of these cases were HIV positive. The impact that HIV will have on the increase of TB in South Africa will largely depend on whether the national TB control programme can improve current cure rates, which are well below the ideal of greater than 80%. In 1996 the cure rate of sputum smear positive pulmonary TB (the form of TB which is most infectious) varied in the different provinces from 33.5% to 65%. This is slowly improving with the implementation of the directly observed treatment strategy. If the dismal cure rates of 1996 are not improved it is estimated that TB will increase approximately threefold

by 2005. If the ideal cure rates are achieved, however, then the increase could be limited to approximately 50% by 2005.

Table 2.5
Annual TB incidence and effect of HIV infection in South Africa (1996)

Province	TB incidence per 100,000	Proportion of HIV+(%)
Western Cape	559	12
Eastern Cape	504	20.4
KwaZulu/Natal	381	45
Gauteng	375	25.2
Northern Cape	340	13.6
Free State	282	32.1
Mpumalanga	286	39.5
North West	271	25.9
Northern Province	260	16.7
Total	362	27

Source : *Epidemiological comments*, October 1996

Tuberculosis incidence and prevalence rates for the Cape Town area are difficult to obtain. Population statistics for many of the townships where tuberculosis is highly prevalent are rough estimates, since the Black population is highly mobile. In 1995, the estimated overall tuberculosis incidence rate was 372/100,000, ranging from less than 25/100,000 for Whites and Asians to approximately 400 and 800/100,000 for Coloureds and Blacks respectively. In certain high-density residential areas in Cape Town, an incidence rate as high as 1339/100,000 has been reported [66]. Of interest though is that the Coloured population has the highest rates (713/100,000 compared with national average of 225/100,000 population in 1993) despite the fact that their socioeconomic conditions have been a bit better than those of Black population [67].

Wood *et al.* have reported a changing pattern in the patient profile, from a predominant homosexual to a heterosexual pattern over an 11-year period in the three main population groups attending their HIV clinics, which are the major HIV referral clinics in Cape Town. This has resulted in significant changes in patient demography and HIV presentation, and an increase in the frequency of both pulmonary and extrapulmonary tuberculosis. They reported a strong interaction between tuberculosis and HIV, with pulmonary tuberculosis as the commonest opportunistic infection in their patients [2]. Drug-resistant tuberculosis was frequently encountered in small number of White male homosexuals. In contrast, HIV infection in the Black and Coloured patient population was not associated with an increased rate of multi-drug resistant tuberculosis [2].

2.4 PATHOGENESIS AND CLINICAL COURSE OF HIV-1

The origin of HIV-1 has been widely debated but still remains an unresolved question. Desrosiers argued that there are two possibilities for the origin of HIV-1: "one possibility is that the virus has always been present in human population, in one form or another, but may have gone unrecognized due to an extremely low prevalence and/or its confinement to isolated population. In this model, features of modern society, urbanization of populations in developing countries, and extensive world travel would have introduced the virus into developed countries. The virus would then be spread by sharing of drug paraphernalia and sexual promiscuity. Alternatively, HIV-1 may have entered human population from another species relatively recently. Increasing prevalence and high mortality would then be consequences of infection of the new host to which the virus was not fully adapted." They argued further that " The

most likely possibility is a combination of these two : that the virus has been entering the human population from an animal reservoir (through the butchering of monkeys, for example) but has been unable until recently to spread from initial limited foci of infection" [68].

HIV-1 can infect a wide range of cells. These include B cells, natural killer cells, precursor CD4+ bone marrow cells, and CD8+ T cells [69].

HIV-1 exhibits strong tropism for CD4+ cells, including lymphocytes and macrophages, and a capacity to persist and replicate in the face of humoral and cellular immune responses, and is associated with the development of progressive immunologic deterioration [70]. Host factors play an important role in determining rates of disease progression in HIV-1-infected individuals. HIV-1 is able to subvert the host immune system by infecting CD4+ T cells that normally orchestrate immune responses and by inducing the secretion of pro-inflammatory cytokines that the virus can utilize for its own replication [71]. Certain chemokine receptors serve as necessary co-factors for HIV entry into its target. Ligands for these receptors can modulate the efficiency of HIV infection and thus influence on the pathogenesis of HIV disease [71].

The ultimate consequence of infection with HIV is profound immunosuppression that is the result of both quantitative and qualitative abnormalities of the helper/inducer subset of T lymphocytes [72]. The initial pathogenic event in HIV infection is binding of envelope glycoprotein of HIV to the CD4 receptor molecule present on the surface of CD4+ T cells and monocyte/macrophages [72]. It has been shown that the CD4

which is present on the surface of CD4+ T lymphocytes and cells of the monocyte/macrophage lineage are the primary receptor for HIV-1 [72]. *In vivo* the reservoir for HIV infection in the peripheral blood is the CD+ T cell, whereas in other tissues the monocyte/macrophages may play a substantial role. As disease progresses in HIV-infected individuals, the viral burden in the peripheral blood CD4+ T cells increases [72].

However CD4+ T lymphocytes and macrophages differ in several important ways. HIV-1 infection of CD4+ T cells *in vitro* leads to extensive cell death, whereas HIV infection of monocyte/macrophages produces a more limited cytopathicity *in vitro* [73]. In CD4+ cells, virions are released almost exclusively at the plasma membrane, whereas in monocyte/macrophages, HIV-1 particles are often released within intra-cytoplasmic vacuoles. CD4+ T cells and monocyte/macrophages also differ in their ability to sustain replication of particular HIV isolates [73]. In contrast to the situation with macrophages, HIV requires that CD4+ T lymphocytes be activated for optimal replication [74,75]. It has been demonstrated that there is a little or no replication of HIV in lymphocytes in resting peripheral blood mononuclear cell cultures (PBMC)[74]. Since most lymphocytes in the blood are in a resting state, only a small fraction is a suitable target for infection. However, both lymphocytes and macrophages are targets of HIV-1 in tissues [74]. Certain tissue-specific disease manifestations of HIV infection such as encephalitis [76] and interstitial pneumonia [77] are associated with extensive infection of the macrophages.

Productive infection of human T lymphocytes by HIV-1 is dependent upon proliferation of the infected cell. Non-proliferating T cells can be infected by HIV-1

and harbour the virus in an inactive state until subsequent mitogenic stimulation. Mitogenic stimulation of infected quiescent cells induces re-initiation of DNA synthesis from partial reverse transcripts. It was shown that HIV-1 DNA synthesis is initiated in infected quiescent T cells at levels comparable with those of activated T cells [78]. The ability of HIV to replicate in the CD+ T lymphocyte and mononuclear phagoctes is strongly influenced by immuno-regulatory cytokines [71]. These cytokines play a role in the regulation of HIV expression in the tissues. The mitogenic signal provided by interleukin-2 (IL-2) leads to both cell proliferation and virus replication in the T-cell [79]. Other HIV-inductive cytokines (*viz.* TNF-α or β) can increase virus expression both in the T-cell or mononuclear phagocytes (MP). It has been shown that the mechanism of action of TNF-α involves the activation of the cellular transcription factor NF-*k*B present in the enhancer region of the HIV proviral LTR. Furthermore, some cytokines (*viz.* IL-1, IL-3, and IL-6) have up-regulatory effects on HIV production. Thus HIV expression is under the control of a complex network of immunoregulatory cytokines [71].

The effect of cytokines *in vivo* in the spreading of HIV remains, however, largely unknown. Cytokines may play a role in tissue damage associated with opportunistic infections and may be involved in the development of many clinical manifestations associated with HIV-1 infection (e.g. HIV-related encephalopathy and in cachexia), and may stimulate the growth of malignant cells in some AIDS-defining conditions (e.g. Kaposi's Sarcoma or lymphomas) [80]. Thus the activation of HIV from latent or chronically infected cells *in vitro* by mitogens and cytokines represents a potential mechanism whereby HIV infection in individuals progresses from an asymptomatic carrier state to clinical AIDS. The release of the virus from activated cells can lead to

the spread of the virus to other target cells and result in both a qualitative or quantitative defect in immunocompetent cells and subsequent immunosuppression [80].

The pathogenic events that occur from initial infection with HIV-1 to the development of clinical disease can better be understood by the schematic diagram below [Fig 2.5] which was proposed by Panataleo *et al.* [81], and Table-2.6 [82].

Fig 2.5
Pathogenic events from primary HIV-1 infection to AIDS

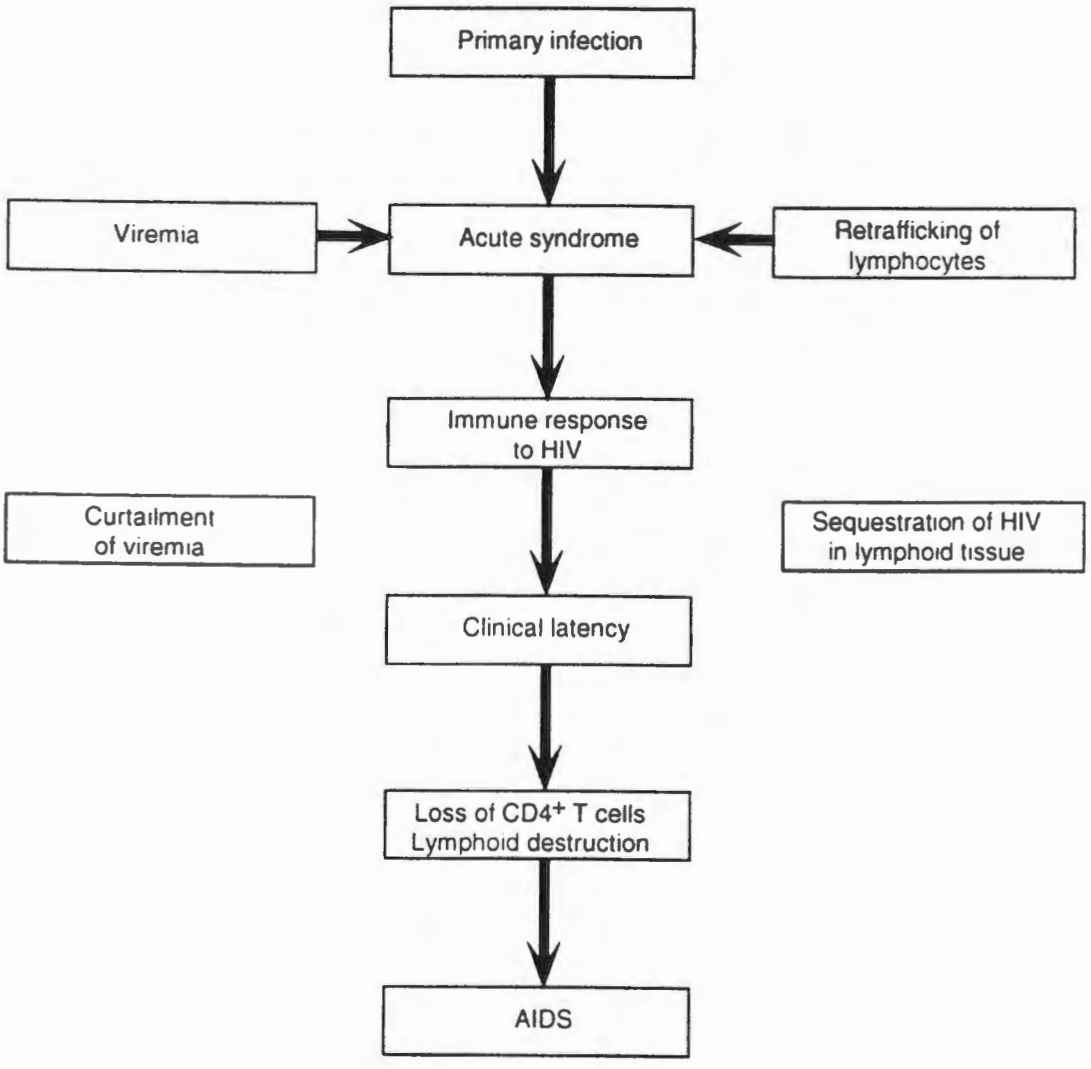


Table 2.6
Pathologic conditions associated with HIV-1

Primary infection

Mononucleosis-like syndrome:

Fever, malaise, pharyngitis, lymphadenopathy, headache, arthralgias, diarrhoea, maculopapular rash, meningoencephalitis

Clinical latency

Often none, but can include:

Fatigue, mild weight loss, generalized lymphadenopathy, thrush, oral hairy leukoplakia, shingles

Clinical disease

200-500 CD4+ T cells/μL:

Generalized lymphadenopathy, oral lesions (thrush, hairy leukoplakia, aphthous ulcers)
Reactivation of herpes zoster (shingles), thrombocytopenia, molluscum contagiosum, basal cell carcinomas of the skin, headache, condyloma acuminata, reactivation of latent *M. tuberculosis*

< 200 CD4+ T cells/μL

Protozoal infections:

Pneumocystis carinii, Toxoplasma gondii, Isospora belli, Crytosporidia, Microsporidia

Bacterial infections:

Mycobacterium avium intracellulare

Fungal infections:

Candida albicans, Cryptococcus neoformans, Histoplasma capsulatum

Viral infections and malignancies:

Cytomegalovirus (CMV), recurrent bouts of oral or genital herpes simplex virus, lymphoma (mostly EBV, some HHV-8),Kaposi's sarcoma (HHV-8), Anogenital carcinoma (HPV)

Neurological symptoms

Aseptic meningitis, myelopathies such as vacuolar myelopathy
Pure sensory ataxia, paresthesia/dysesthesia,
Peripheral neuropathies such as acute demyelinating polyneuropathy, monoeuritis, multiplex, and distal symmetric polyneuropathy
Myopathy
AIDS dementia complex

Tindall C, Cooper DA. AIDS 1991

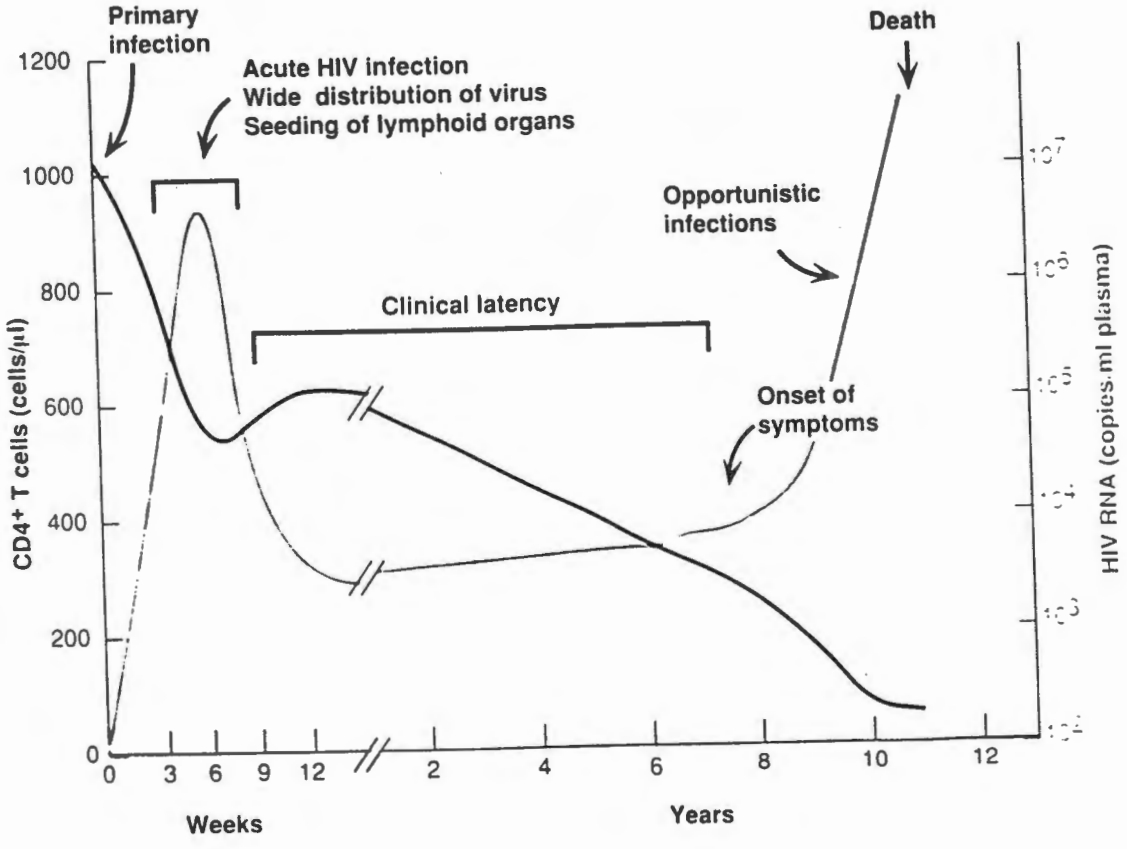
Primary infection of humans with HIV-1 is associated with an acute mononucleosis-like clinical syndrome which appears approximately 3-6 weeks following infection. The severity and persistence of symptoms vary considerably. Significant declines in the levels of CD4+ T lymphocytes in the peripheral blood occur in the first 2-8 weeks following HIV-1 infection [81].

The acute syndrome associated with primary HIV-1 infection is accompanied by a burst of viral replication that can be detected in the blood approximately 3-6 weeks following infection (Table 2.6) [82]. During this period, infectious virus and viral protiens can be readily detected in the cell-free plasma as well as in cerebrospinal fluids, and the number of virions in cell-free plasma can reach 10^6 to 10^7 per milliliter [83].

Aprroximately 3-6 weeks after the infection with HIV-1, specific antiviral immune responses can be detected [82]. These antiviral immune responses are associated with a precipitous decline in plasma viraemia burden in PBMCs, resolution of the clinical syndrome, and a temporary stablization of the number of CD4+ T cells [74]. Both humoral and cellular immune responses can be detected. Multiple factors probably contribute to the decline in viral titer, including the responses of the two arms of the immune system, the secretion of virus-suppressing cytokines, and the possible exhaustion of suitable CD4+ target cells. Koup *et al.* have shown that that the initial decline in plasma viremia is associated with the appearance of HIV-specific, CD8+ cytolytic T cells, whereas the first neutralizing antibody is not detected for several additional weeks [84]. The vast majority of infected individuals develop detectable antibody responses within 3 months of infection [84].

Following the induction of an immune response to HIV-1 in humans, there is usually a relatively long period that is characterised by few clinical manifestations [Fig 2.6]. Clinical symptoms are usually mild. Current estimates for the average time from infection with HIV-1 to the development of the clinical conditions that define AIDS is 9 (range 2-20) years [85]. The time interval from infection to development of opportunistic disease varies greatly from one patient to another [83]. However, the estimated time has gradually increased since AIDS was recognized. This may be due both to better detection of infection and to antiretroviral therapies and effective prophylaxis for life threatening opportunistic infections.

Fig 2.6
Typical course of HIV infection



Pantaleo et al. 1993

Throughout the latent period, there is usually a steady decline in the numbers of CD4+ lymphocytes [Fig 2.6]. The asymptomatic phase may be either much shorter or absent. According to the 1993 revised definition of AIDS by the Centers for Disease Control and Prevention CDC [appendix I], even if the individual is otherwise completely asymptomatic, a diagnosis of AIDS is made if the CD4+ T-cell count falls below 200 per microliter.

2.5 HOST RESPONSE TO TUBERCULOSIS

The causative agent of tuberculosis was discovered by Robert Koch in 1882. Very shortly afterwards, Paul Ehrlich discovered the "acid-fastness" of the tubercle bacillus and introduced a staining technique [88].

Mycobacterium tuberculosis a complex unicellular organism contains a single circular chromosome with many antigenic proteins and sugars. There are two types of antigens: cytoplasmic (soluble) and cell-wall lipid-bound (insoluble) antigens. The soluble antigens can further be divided into those that remain within the cell cytoplasm until released by autolysis or mechanical disruption and those that are actively secreted by living whole cells [89].

Pathogenicity (*patho*: a Greek word meaning disease) has been defined as the quality of a micro-organism of being capable of causing disease; a quality that varies in degree [90]. This property depends on the susceptibility of the host as well as the aggressiveness of the invading organism. The pathogenicity of *M. tuberculosis* is primarily due to its ability to penetrate and survive within the macrophages [91]. Cell-mediated immunity is the defense mechanism which the targeted cell uses to prevent

M. tuberculosis penetration. This cell-mediated immune response to *M. tuberculosis* serves two purposes: either to enhance the ability of the macrophage to inhibit or destroy *M. tuberculosis* or to detect macrophages and other cells that are not controlling intracellular bacillary replication and to lyse them [92]. This may results in releasing *M. tuberculosis* such that it can be engulfed by immunologically effective phagocytes. Dannenberg has shown that the products of cell lysis inhibit *M. tuberculosis* growth and generate an environment that is deleterious to *M. tuberculosis* growth and survival [93].

The immunological phenomena against *M. tuberculosis* comprise three steps: (1) recognition (2) response and (3) reaction to *M. tuberculosis*. *M. tuberculosis* recognition is a property of lymphocytes [92]. The lymphocytes are divided into two main sets: the B-cells and the T-cells. The former mature into anti-*M. tuberculosis* secreting plasma cells, while the latter are involved in the initial recognition of *M. tuberculosis*, the regulation of the immune response by secretion of hormone-like chemical messengers called cytokines and in cell-mediated cytotoxity [92].

The T-cells are divisible into subsets according to their functions and by the presence of certain markers; namely, the cluster differentiation (CD) antigens detectable by means of monoclonal antibodies. The principal T-cell subsets are those bearing either the CD4 or CD8 markers. The former includes the helper and inducer cells while the latter includes suppressor and cytotoxic cells. The activities of the different subsets of T-cells are largely due to the cytokines that they produce [94]. A number of well characterised cytokines produced by cells of the immune system are termed interleukins. Mosmann and Moore have shown that CD4+ T-helper (TH) cells mature

along separate pathways, resulting in cell populations with quite different functions [95]. These are recognised by the nature of cytokines that they secret and are termed TH1 and TH2. Helper T-cells are also divisible according to differences in the structure of the receptor molecules that bind to *M. tuberculosis*. These receptors consist of two protein chains which, in most cells, are of types termed alpha and beta but a minority of these cells have gamma and delta chains. The latter, termed gamma-delta (γ-δ) cells are usually CD4-/CD8- but a minority are CD8+ [95].

Each lymphocyte bears a receptor that binds to just one of the thousands of possible *M. tuberculosis* determinants or epitopes. There are many sub-populations of lymphocytes, each specific for just one epitope. Small numbers of *M. tuberculosis*-specific lymphocytes proliferate to form clones of cells of sufficient numbers to mediate an effective immune response [96]. Before *M. tuberculosis* is able to induce such clonal proliferation it must be presented to the lymphocytes in a special way. This is the task of the antigen presenting cells (APCs) which include cells of the monocyte/macrophage series, dendritic cells of lymph nodes and scattered lymphoid tissues and the Langerhans cells of the dermis. Other cells may, under certain circumstances, also serve as APCs. The APCs engulf *M. tuberculosis* which contain a multitude of epitopes [97]. Some of these epitopes are actively secreted and some are somatic, and only released within lysosomes.

M. tuberculosis epitopes are presented on the surface of the APC in close relation to host molecules which are products of the major histocompatibility complex (MHC) [98]. There are two types of such molecules: Class I, and class II. The Class II molecules are principally found on cells with specific *M. tuberculosis* presenting

functions and activate the CD4+ helper/inducer T-cells. Class I molecules are present on all cells and activate the CD8+ suppressor/cytotoxic T-cells [99]. Binding of *M. tuberculosis*-specific lymphocytes to the epitope/HLA complex causes a signal to be delivered by the APC to the lymphocyte, inducing the latter to secret interleukin-2 (IL-2) which mediates lymphocyte division and clonal expansion. Epitopes from *M. tuberculosis* within phagosomes are presented by MHC Class II molecules while those from *M. tuberculosis* that lie freely in the cytoplasm of the cell are presented by class I molecules. This difference in presentation enables the immune system to decide whether to enhance the microbicidal power of the cell or to destroy it and thereby liberate its contents [99].

Macrophages belong to the same cell lineage as the blood-borne monocytes. Macrophages are phagocytic cells, but, unlike the other major class of phagocytes, the polymorphonuclear leukocytes, they are long-lived cells which settle in a given tissue or organ and adapt to the local environment. Thus, alveolar macrophages, osteoclasts, Kupffer cells of the liver and Schwann cells of the nerves are all specialized macrophages. Macrophages do not express their full antimicrobicidal potential unless they are activated. Such activation is mediated by gamma interferon (IFN-γ) which is secreted by the expanded population of CD4+ helper T-cells [100].

In addition to its microbicidal activity, the macrophage synthesizes and secretes many important compounds that affect the pathogenesis of *M. tuberculosis*. These include some of the acute-phase reactant proteins, and protease that liquefies necrotic tissue and thereby contributes towards a cytokine termed tumour necrosis factor (TNF-α) [101]. TNF-α is an important mediator of tissue-necrotizing immunopathology. The

systemic release TNF-α is responsible for extreme wasting with advanced tuberculosis [102].

An important characteristic of chronic tuberculosis is the formation of the granuloma. Macrophages in granulomas are termed epithelioid cells. Many cytokines, including TNF-α, are involved in granuloma development and regulation [103].

In order to survive within a macrophage, the *M. tuberculosis* must be able to resist destruction by the wide range of killing mechanisms of the infected cell. These mechanisms include enzymes such as lysozyme, lipases and phosphatases. The process of phagocytosis begins by engulfing *M. tuberculosis* by the cell membrane. The phagosome then fuses with the lysosomes which contain the bactericidal agents [103]. *M. tuberculosis* can survive within the phagosome either by inhibiting the phagosome/lysosome fusion, or by covering itself with a protective layer that absorbs or neutralizes the bactericidal agents or, it may escape from the vesicle and lie freely in the cytoplasm of the cell [103]. *M. tuberculosis* is able to escape from the phagosome and replicate in the cytoplasm [104]. This happens when the cell becomes immunologically effete and unable to control intracellular growth or when *M. tuberculosis* enters cells other than the macrophages [104].

If the macrophage cannot control *M. tuberculosis* growth, it is killed with release of the *M. tuberculosis* which initiates an inflammatory response which, in turn, attracts blood-borne phagocytes and other white cells, including natural killer cells and γ-δ T-cells. The latter cells recognize certain *M. tuberculosis* components and cause the formation of a low-turnover ganuloma of the *M. tuberculosis* type. This to some

extent limits the progression of the disease process before the development of the specific immunity mediated CD4+ T-cells. Early recognition of *M. tuberculosis* by γ-δ cells is required for subsequent activation of antigen-specific α-β CD4+ T-cells [104,105].

However, protective cell-mediated reactions are not the only immune phenomena associated with *M. tuberculosis*. Others, such as immunosuppression and delayed hypersensitivity [106], also occur and make the process more complex. These other reactions are of great relevance to the pathogenesis of the disease.

2.6 EFFECT OF THE HOST RESPONSE TO TUBERCULOSIS ON HIV-1 PATHOGENECITY AND DISEASE PROGRESSION

In immunocompetent individuals, following aerosol infection and phagocytosis by pulmonary macrophages, *Mycobacterium tuberculosis* will elicit a cell-mediated immune reaction in lung tissue and regional lymph nodes. CD4 T-lymphocytes are important regulators of the induced immune response [102]. Cell-mediated immunity involves an incompletely understood interplay between TH1 CD4+ T-cells that produce interferon-γ and interleukin (IL)-2, cytokine-activated macrophages that produce TNF-α, IL-1 and IL-6, and cytotoxic CD4 T-cells [103,104]. Cell mediated immunity leads to granuloma formation at the site of infection, and macrophage activation results in killing of phagocytosed mycobacteria. Delayed hypersensitivity, an important mechanism for clearing mycobacteria that escape killing by cell-mediated immunity, acts by macrophage lysis.

Although the cellular immune activation of T cells and macrophages in response to tuberculosis is beneficial to the host in terms of the control and killing of *Mycobacterium tuberculosis*, it is deleterious in terms of HIV-1 disease. Among immune responses to *Mycobacterium tuberculosis*, a number of cytokines are produced in large amounts in affected organs [71]. Several of these cytokines have *in vitro* up-grading (TNF-α, IL-6) or bifunctional (IL-10, IFN-γ) activities on HIV replication. This cytokine network, which is involved in the regulation of normal immune responses, may play a role in the pathogenesis of human immunodeficiency virus (HIV) infection, by altering the replication of HIV in target cells. In vitro data suggest that certain cytokines like TNF-α, granulocyte-macrophage colony-stimulating factor (GM-CSF) and interleukin-6 (IL-6) could up-regulate HIV expression in T-cells and macrophages [69,71,78,79,81].

Several studies have suggested that tuberculosis may increase the ability of HIV to replicate in the CD4+ T cells by activating T cells and macrophages that may harbour latent HIV-1. The extent to which HIV-1 replication is altered following the cellular activation induced by tuberculosis has been documented in many studies.

In a study from Uganda, Wallis *et al.* found that antigen-induced blastogenesis (a phenomenon that increases with stage of HIV infection, decreases in response to antiviral therapy, and predicts progression to AIDS) and production of TNF-α were 3-10 times greater in HIV-1-seropositive patients with pulmonary tuberculosis compared to HIV-1-positive controls without tuberculosis. They argued that this result was "most likely a consequence of the cell-mediated cytotoxicity directed against *M. tuberculosis*-infected macrophages" [107].

Garrait *et al.* assessed the role of pleural fluids recovered from seronegative patients with TB on HIV replication in acutely infected blast cells. They observed that tuberculous pleural fluids stimulated HIV replication *in vitro.* TNF-α, IL-6, IFN-γ, and granulocyte-macrophage colony-stimulating factor (GM-CSF) were detected in tuberculous pleural fluids. They observed further that anti-TNF-α and anti-IL-6 Abs decreased HIV replication by 60% and 90%, respectively, and that unstimulated tuberculous pleural lymphocytes were productively infectable by HIV [108].

In addition, it has been shown that the onset of *Mycobacterium tuberculous* disease in HIV-patients results in a significant increase in plasma HIV load. One study suggested that antigen–specific activation of HIV-infected CD4 cells by *Mycobacterium tuberculosis* enhances viral replication and makes CD4 more vulnerable to HIV infection. In a study by Goletti *et al.,* in which plasma viral load was measured in HIV-infected individuals before, during, and after the development of TB; a 5- to 160-fold increase in viral replication was observed during the acute phase of MTB disease. Their study demonstrated that *Mycobacterium tuberculosis* induced HIV replication in CD8+ T cell-depleted lymphocytes from HIV-infected individuals with a history of purified protein derivative (PPD) positivity but not in those who were PPD negative. Furthermore, this induction of HIV replication was observed to correlate with the level of cellular activation. In their *in vitro* acute HIV infection model, *Mycobacterium tuberculosis* increased HIV replication in PBMC from healthy donors with a history of PPD positivity, but not in PBMC from PPD-negative donors. This induction of viral replication also correlated with cellular activation. They concluded that "*Mycobacterium tuberculosis* increased HIV replication both *in vivo* and in *in vitro*. This *Mycobacterium tuberculosis*-mediated

viral production likely occurs through antigen-specific activation and infection of responding T cells" [109].

Mycobacterium tuberculosis was also reported to enhance HIV-1 replication by transcriptional activation. Zhang *et al.* found a striking increase from 4- to 208-fold in p24 levels in bronchoalveolar lavage fluid from involved sites of *Mycobacterium tuberculosis* infection *vs* uninvolved sites in three HIV+ patients. They used an *in vitro* cell culture model to determine if tuberculosis could activate replication of HIV-1. Mononuclear phagocyte cell lines U937 and THP-1 infected with HIV-1JR-CSF, *in vitro* and stimulated with live *M. tuberculosis* H37Ra, had a threefold increase in p24 in culture supernatants. Using the HIV-1 long terminal repeat with a chloramphenicol acetyltransferase (CAT) reporter construct, they have shown that *M. tuberculosis* increased transcription 20-fold in THP-1 cells, and that cell wall components stimulated CAT expression to a lesser extent. Furthermore, they observed that antibodies to TNF-α and IL-1 inhibited the increase in CAT activity of the HIV-1 long terminal repeat by *M. tuberculosis from* 21-fold to 8-fold. They argued that "stimulation of HIV-1 replication by *M. tuberculosis* may exacerbate dysfunction of the host immune response in dually infected individuals" [110].

Similar findings were observed in SIV/mac-infected monkeys. Zhou *et al.* [111] have explored T cell responses to AIDS viruses using a SIV/mac animal model. In their study, T cell activation correlated with a marked increase in viral loads in SIV/mac-infected monkeys. In addition they observed a prolong T cell activation which coincided with an enhanced decline of CD4+ PBL counts and an accelerated progression to clinical AIDS in the co-infected monkeys. Within 2 to 7 months after

BCG coinfection, all chronically SIV/mac-infected monkey died from SIV induced AIDS including tuberculosis-like disease. They argued that "*Mycobacterium*-driven T cell activation may be the mechanism underlying the enhanced pathogenicity of AIDS virus infection in the coinfected individuals" and that the results of their model "strongly support the hypothesis that active coinfection with HIV and *Mycobacterium* can impact remarkably on the AIDS virus-induced disease" [111].

In brief, *Mycobacterium tuberculosis* permits an enhanced HIV replication at the site of mycobacterial infection with a subsequent increase of viral load in HIV-infected subjects. The cytokines produced during the immune response to Mycobacteria might result in both activation of latently infected cells and induction of virus expression, as well as in an increased replication from infection of newly activated cells. In addition, *Mycobacterium tuberculosis* partially suppresses factors responsible for controlling HIV replication such as IL-10 [95]. These factors combined could participate in an increased viral burden subsequent to the development of tuberculosis and might partially explain HIV-1 disease progression following the onset of tuberculosis. These findings may explain not only why HIV progresses more rapidly in regions where much of the population is chronically infected with *Mycobacterium tuberculosis*, but also why HIV seems to be more easily transmitted within these populations.

2.7 THE CLINICAL IMPORTANCE OF TUBERCULOSIS IN HIV INFECTION

Given the aforementioned facts, tuberculosis is thus of a particular importance in HIV-1 infection. In addition, tuberculosis is frequently found in HIV-infected patients and usually occurs in HIV-infected patients without pre-existing AIDS; occurring in

50%-67% of HIV-infected patients [111]. This is presumably because *Mycobacterium tuberculosis* is more virulent than other HIV-associated pathogens, such as *Pneumocystis carinii* pneumonia and *M. avium* complex [112]. Tuberculosis is therefore more likely to cause disease at an early stage of immunodeficiency and to provoke poor prognosis [113-115].

An important clinical implication of the above is that delayed diagnosis of tuberculosis in patients with HIV infection could lead to a significant increase in mortality. Of 52 such patients with tuberculosis evaluated at one medical center, 25 (48%) either died with untreated tuberculosis or began to receive antituberculosis therapy more than three weeks after presentation. Forty-five percent of those whose diagnosis was delayed died of tuberculosis, as compared with 19 % of those in whom the diagnosis was made rapidly, suggesting that delayed diagnosis results in increased mortality. Delayed diagnosis was not due to atypical manifestations of tuberculosis but due to errors in management in 84% of the patients [116].

The diagnosis of tuberculosis may thus permit early identification of HIV infection, the institution of antiretroviral therapy and counselling to reduce both the transmission and the pathogenecity of the virus.

2.8 CLINICAL DIAGNOSTIC FEATURES OF HIV- RELATED TB

Similar to tuberculosis in HIV negative patients, the lung is the commonest site of tuberculosis in HIV infection. Pulmonary tuberculosis may occur at all stages of HIV infection, occurring in 42-80 % of cases in Africa, and 77 % of cases in Brazil [57,117,118]. The clinical and radiographic presentation of pulmonary tuberculosis,

and the diagnostic yield of smears, however, varies with the degree of immune compromise. The most common features accompanying the presentation of HIV-associated tuberculosis in Africa are oral candidiasis, generalized lymphadenopathy and recent history of herpes zoster [43,51]. In the USA, sputum smears have been found to be positive in 31-82% of HIV-seropositive patients with pulmonary tuberculosis, with the higher fractions in less immunocompromised individuals (those with typical reactivation tuberculosis) and the lower fractions in advanced HIV disease [119]. In developing countries, rapid diagnosis of pulmonary tuberculosis is based on simple diagnostic techniques: sputum smear microscopy and chest radiography, when available. In a series of 62 HIV-positive patients from Zambia sputum smears were positive in 63% of culture proven pulmonary tuberculosis cases, but showed a low number of bacilli (defined as 1 to 10) in 35%, as compared to 82% and 25%, respectively, in 61 HIV-seronegative patients [51].

The tuberculosis detection rate based on sputum smear examination may thus be comparable to that described for HIV-seronegative patients, at least during the early stage of HIV disease. On the other hand, when the immune system is severely compromised, the sensitivity of sputum smear examination may be reduced [120]. However, it has been observed that even in absence of cavitation and without liquefaction necrosis, the profound impairment of the immunity seen in HIV infection may permit tubercle bacilli to multiply in such large numbers as to become visible on smear examination [119,121].

In a radiological investigation of 963 HIV-infected adults at three central African hospitals, Tshibwa-Tuma *et al.* showed that HIV-infected group of patients with

tuberculosis had a significantly higher proportion of lymphodenopathy than HIV-negative TB patients (26% *vs* 13%; $p = 0.001$), pleural effusions (16% vs 6.8%; $p = 0.001$), miliary shadowing (9.8% vs 5%; $p = 0.001$), an interstitial pattern (12% vs 7%); $p = 0.01$) and consolidation (10% *vs* 3%; $p = 0.001$). There was significantly less cavitation (33% *vs* 78%; $p = 0.001$) and atelectasis (12% *vs* 24%; $p = 0.001$) seen in the HIV-positive group compared to the HIV-negative group of TB patients. These patterns of radiographic changes were consistently seen across all three hospital sites [122]. In Uganda, chest X-ray findings not typical of tuberculosis were observed in 41 % of HIV-seropositive TB patients as compared to only 16% of HIV-negative TB patients [56].

Cavitation is less common in HIV-infected patients with tuberculosis and may range between 16% and 48%, whereas usually more than half of the immunocompetent patients develop pulmonary cavities [43,52]. The lack of cavitating lesions in HIV-infected patients probably depends on the host's altered immune response; an abnormal interaction between host and bacillus results in an outcome different from the expected [52]. Upper lobe involvement is seen less frequently in HIV-seropositive patients, whereas hilar or mediastinal lymphadenopathy is common [43,52]. In a study of tuberculosis in Haiti involving 10 patients with AIDS, 57 HIV-seropositive patients without AIDS, and 158 seronegative patients, a post-primary pattern was seen in 20%, 51% and 80% of patients respectively, whereas an atypical primary-like pattern was seen in 80%, 30 % and 11% respectively, which supports the observation that the more advanced the stage of disease, the more common the atypical presentation. In this study cavities were significantly less common in HIV-seropositive patients, while hilar or mediastinal adenopathy without parenchymal infiltrates was more common

[121]. In Brazil, an atypical chest X-ray was reported from 81% of AIDS patients as compared to 6% of non-HIV infected persons. Similarly, observations from the USA suggest that the chest radiographic manifestations of HIV-related pulmonary tuberculosis reflect the stage of HIV infection, with a typical reactivation pattern early in the course, and an atypical one in the more advanced stages [118].

Table 2.7

Estimated frequency of chest radiograph findings in patients with tuberculosis with and without HIV infection in developing countries

Chest radiograph Finding	HIV (+) Patients (%)	HIV(-) Patients (%)
Typical pattern[a]	46-59	80-85
Atypical pattern[b]	40-81	6-16
Upper lobe infiltrates	50-69	72-90
Cavitations	16-48	35-87

(Bulletin of WHO 1992:72)

a Post-primary pattern : upper lobe infiltrates, cavitations.
b Primary-like pattern : intrathoracic adenopathy , with or without parenchymal disease , localized non-cavitary disease in middle or lower lobes , isolated pleural effusion or no abnormalities.

Extrapulmonary tuberculosis is a common finding among HIV-infected patients in developing countries. Available information shows that HIV-seroprevalence is higher in patients with extrapulmonary disease than in those with pulmonary disease alone [43,51]. In Africa during the pre-AIDS era only a small fraction of cases occurred with extrapulmonary or combined pulmonary and extrapulmonary features [123-124]. A change in the distribution has occurred in recent years. In a series of HIV-related tuberculosis cases from the Central African Republic 31% had at least one extrapulmonary site of disease [124]. In two other series from Malawi and Kenya, 57% and 53%, respectively, of HIV seropositive patients had extrapulmonary

tuberculosis, compared to 20% and 19% of HIV seronegative patients [117,124]. In the USA, one study has shown that only 38% of tuberculosis diagnosed in patients with advanced HIV infection occurred in the lung alone, while 30% were entirely extrapulmonary and 32% both pulmonary and extrapulmonary [125].

Lymphadenopathy, both intra-and extrathoracic, remains the most frequent form of extrapulmonary tuberculosis reported from the Central African Republic, Zambia, Kenya, and Brazil [43,51,118,126].

Disseminated miliary forms of TB are also frequent in HIV positive patients [17,45]. However, mycobacteraemia may be difficult to diagnose in developing countries in the absence of routine blood cultures [45,52,120] Virtually all organs have been reported as extrapulmonary sites of tuberculosis, including the gastrointestinal tract, liver, meninges, genito-urinary tract, peritoneum, and bone marrow [15,45,47].

2-9 SURVIVAL OF PATIENTS WITH HIV-RELATED TUBERCULOSIS

The HIV epidemic has had a major impact on the incidence, mortality and the case fatality rate of tuberculosis. It is unclear, however, whether tuberculosis affects the course of HIV disease, and there is sparse and insufficient evidence on whether tuberculosis shortens survival in patients with HIV infection. One review concluded that there was no noticeable decrease in survival attributable to tuberculosis in patients with HIV infection [125]. The authors of this overview may have based their conclusion on the following. Firstly, in patients who had died with both AIDS and tuberculosis the cause of death was almost invariably attributed to AIDS and not to

to active tuberculosis. However, the validity of a cause of death diagnosis may be limited when there are several possible causes of death. Secondly, studies from Brazil [126], Spain [128], and Europe [129] have indicated that patients with tuberculosis as the AIDS defining disease survive longer than other AIDS patients. Some authors of these studies argued that since clinical tuberculosis may be manifested earlier than other AIDS defining diseases in HIV infection, the apparently longer survival in these patients may be due entirely to earlier diagnosis of AIDS [126].

However, different findings from those of the reviews cited above have been documented in other studies. In HIV-infected patients, some studies have suggested that survival is shorter in patients with active tuberculosis than in patients without active tuberculosis [130,131]. This higher mortality occurs despite adequate response to antituberculosis therapy in most cases.

2-10 SURVIVAL OF PATIENTS WITH HIV-RELATED TUBERCULOSIS IN DEVELOPED COUNTRIES

So far, the major studies assessing the role of tuberculosis in HIV progression have been reported from developed countries. Whalen *et al.*, in a retrospective cohort study at four U.S. medical centres comparing the survival (and incidence rate of opportunistic infections) in 106 HIV-infected patients with active tuberculosis ("cases") with that of 106 HIV-infected patients without tuberculosis ("controls"), reported an overall shorter survival among cases than controls. Active tuberculosis was associated with an increased risk of death during the follow-up period (odds ratio = 2.17) even after controlling for confounding [130].

Furthermore, the incidence rate of new opportunistic infections in active tuberculosis subjects was 4 infections per 100 person-months compared with 2.8 infections per 100 person-months in subjects without TB (incidence rate ratio = 1.4, 95% confidence interval [CI] = 0.94-2.11). They concluded that "although active tuberculosis may be an independent marker of advanced immunosuppression in HIV-infected patients, it may also act as a cofactor to accelerate the clinical course of HIV infection" [130].

In France, in a similar study of HIV infected subjects, Leroy *et al.* compared survival of 104 "cases" with active tuberculosis with 620 "controls" without active tuberculosis [131]. They reported an increased mortality in cases compared to controls (risk ratio = 1.5, 95% confidence interval = 1.2-2.1) even after controlling for differences at entry. In this study the risk of AIDS-defining opportunistic infection or a decrease to fewer than 50 CD4+ cells/mm^3 was slightly but not statistically greater in tuberculosis patients than in controls. Tuberculosis disease thus reduced survival but did not affect the occurrence of subsequent opportunistic infections or rate of CD4+ count decline.

LIMITATIONS OF THESE STUDIES :

1. To assess the role of tuberculosis disease on the course of HIV infection, the ideal study design would be to compare HIV disease progression in tuberculosis patients and patients without tuberculosis in an incident cohort followed prospectively from HIV seroconversion. The retrospective nature of the above studies raises the question of accuracy and completeness of the data and precluded the collection of potentially important variables. For example, in the first study the authors reported that chest radiograph interpretations were available for analysis on only a subset of the study subjects.

2. The case definition used in these studies systematically excluded subjects with culture-negative tuberculosis. If this exclusion was associated with mortality, and if culture negative tuberculosis cases have lower mortality, then a selection bias may have occurred. This may, partially or completely, explain the observed results of a poorer survival of tuberculosis patients and may preclude the generalizability of the findings of these studies to patients with tuberculosis diagnosed differently.

3. It was observed in these studies, and in other studies of relevance from Western countries, that most of the HIV-infected patients were predominantly intra-venous drugs users. There has been a suggestion of higher mortality in injecting drug users with HIV, arising from drug overdose and violence, including homicide and suicide. In addition, compared with other patients acquiring HIV infection through other modes of transmission, they have been reported to experience a faster rate of CD4 lymphocyte decline, which could explain a shorter survival time in this group. Whalen *et al.* argued that assuring compliance with primary tuberculosis prophylaxis or complete and regular follow-up is challenging in such patients [130]. If the proportion of such patients is not the same among "cases" of active tuberculosis disease as among "controls" without active tuberculosis disease, and if " controls" preferentially received antiretroviral therapy and tuberculosis prophylaxis, or are more compliant with prescribed therapy, the beneficial impact of therapy alone may account for differences in survival.

4. The above two studies produced conflicting results regarding the occurrence of opportunistic infections. The Leroy study [131] did not show an accelerated progression of HIV related opportunistic infection in relation to tuberculosis disease. In contrast, the findings of the Whalen study [130] indicated greater, though not significant, occurrence of opportunistic infections in tuberculosis patients than in controls. This increased occurrence of opporunistic infections among tuberculosis patients could, however, be partially or wholly attributable to detection bias where tuberculosis patients may have been followed more closely than controls.

5. It is presumed that more than 90% of tuberculosis cases that occur each year in the western countries are the result of remote rather than recent infection with *Mycobacterium tuberculosis* [123]. Reports of AIDS-associated tuberculosis from western countries suggest that remote infection might be responsible for the development of disease in HIV-infected patients. This fact may complicate determining with certainty the pathogenesis of tuberculosis in HIV/AIDS-infected patients from the western countries. In western populations, the prevalence of pre-existent tuberculosis infection is low, and usually most of the patients develop tuberculosis after AIDS is diagnosed, unlike patients from developing countries.

Whilst the annual incidence of tuberculosis fell to around 10/100,000 in some developed countries, it remained at over 200/100,000 in sub-Saharan Africa before the advent of HIV [133]. The annual risk of infection with *Mycobacterium tuberculosis* in sub-Saharan Africa remains extremely high;

at over 1% per year in some large surveys and probably even higher in poor areas, indicating that tuberculosis has never been adequately controlled in these countries. This implies there may be a differential immunity against TB between HIV-infected patients from developed countries, where TB is uncommon, and those from developing countries where most patients are exposed to TB. Furthermore, most deaths in developed countries have been reported to occur from complications of HIV disease and not directly from complications of tuberculosis, a result which may be limited to patients from developed countries. By contrast, active tuberculosis has been reported as the commonest cause of death in HIV-infected patients in African countries [51,52,53,55,65,61,62]. These differences might have some implications for the differences reported in survival after the diagnosis of active tuberculosis in patients in the two settings.

6. These studies included only patients with both tuberculosis and AIDS as defined by the CDC surveillance criteria, by which the AIDS definition was expanded to include tuberculosis as an AIDS-qualifying condition. By disproportionately focusing on cases of tuberculosis occurring in patients with AIDS, an advanced stage of HIV infection, these retrospective analyses may have underestimated the frequency but overestimated the severity of tuberculosis as an HIV-related illness.

In the most comprehensive prospective study based on a large population cohort of European AIDS patients from 52 centres in 17 countries, in which 5249 patients who were alive and free of tuberculosis one month after diagnosis of AIDS, enrolled

between 1979 and 1989, followed up untill 1992, a seemingly paradoxical result occurred [127]. Patients who developed active tuberculosis at some point during their follow-up survived longer after their diagnosis of AIDS than patients who remained free of tuberculosis. However, the authors argued that the observed results may alternatively be explained by other factors that were not measured, such as severe immune suppression or malnutrition that may have induced both active tuberculosis and death in AIDS patients.

Similarly, in a most recent study done in the USA, Munsiff *et al.* found that tuberculosis was not an independent predictor of increased non-tuberculosis-related mortality in HIV-seropositive. The non-significant association between TB and mortality in that prospective study was observed despite the fact that TB was shown to be an independent predictor of subsequent AIDS-defining illness (RR, 4.1; 95% confidence intervals 1.9-8.7) [134].

2-11 SURVIVAL OF PATIENTS WITH HIV-RELATED TUBERCULOSIS IN DEVELOPING COUNTRIES

The association of tuberculosis with the immunodeficiency that was later recognized as HIV infection was first recorded among Haitian immigrants to the USA [113], but it rapidly became clear that tuberculosis was also a major feature of HIV infection in Africa. Most of the research work on the effect of HIV on tuberculosis in developing countries comes from studies in Zaire, Cote d'Ivoire, Kenya, Zambia and Tanzania. It is difficult to obtain clear evidence from these studies or to pool the results of the survival of HIV infected patients with active tuberculosis in one representative estimate owing to the following :

1. Most of these studies focused on the contribution of HIV infection to the apparently increasing incidence of tuberculosis in central Africa, and the redirection of public health priorities towards tuberculosis as the major treatable and preventable complication of the AIDS epidemic [43,45,47,51,52,55,56,57]. Only few of these studies have addressed the question of the prognostic effects of tuberculosis in HIV progression. Furthermore, most of these few studies have compared the survival of HIV positive and negative patients with tuberculosis, rather than comparing survival of HIV positive patients with and without active tuberculosis. In other words, the major concern of these studies was the effect of HIV infection on the incidence and natural history of tuberculosis, rather than vice versa.

2. In the studies mentioned above, only patients with smear positive tuberculosis were included. The diagnosis of tuberculosis in these studies depended heavily on microscopy of the sputum smear. However, studies from USA and Europe have cast some doubt on its sensitivity. As cited in the last section, the rate of concentrated sputum smear positivity in HIV positive patients with pulmonary tuberculosis confirmed by culture is very low compared with HIV negative patients. Indeed For every case of sputum-smear-positive tuberculosis, there are many cases of sputum-smear-negative and extrapulmonary tuberculosis. Direct measurements of the concentration of viable tubercle bacilli in sputum confirm that HIV positive patients excrete significantly fewer organisms/ml sputum than HIV negative patients with culture confirmed pulmonary tuberculosis [53,62]. It has been reported that nearly half of HIV positive patients with tuberculosis will have no clinical features specifically suggestive

of tuberculosis [53,61,62]. The systematic exclusion of smear negative tuberculosis patients from these studies may have introduced a bias towards not including those with more advanced immunodeficiency who are likely to be smear negative. This bias may be responsible for the observed results in studies from developing countries.

3. Rifampicin in HIV-infected patients with TB has been shown to improve early mortality [55]. In addition to its anti-mycobacterial action, some authors argued further that rifampicin has a broad-spectrum antibacterial activity as well. In African countries this intervention, which is relatively expensive, is not as widely used as compared to developed countries. This, to some extent, may explain the poor prognosis of HIV-infected patients with TB in Africa.

From the above discussion, it is clear that the evidence for the role of tuberculosis in HIV progression gained so far from studies conducted in developed and developing countries is inconsistent and somewhat inadequate. As discussed above, factors such as the wide spectrum from which the patients of these different studies were drawn, application of non-comparable and different techniques and methods in patient recruitment, follow-up and diagnosis, and other differing factors such as HIV subtype, exposure to endemic diseases, nutritional status, and access to medical treatment, might be responsible for the inconsistent and contradictory results. It is apparent that further HIV positive cohort studies are needed, to delineate the sequence of immunologic events before and immediately after tuberculosis and to explore the mechanism of interaction between HIV and tuberculosis.

2-12 VALIDITY OF HIV/AIDS STAGING SYSTEMS IN THE AFRICAN SETTING

The CDC revised the classification system for HIV infection [37] to emphasize the clinical importance of the CD4+ T-lymphocyte count in the categorization of HIV-related clinical conditions. This classification system, which was introduced in 1993, replaced the system proposed by CDC in 1986. Consistent with the 1993 revised classification system, CDC has also expanded the AIDS surveillance case definition to include pulmonary tuberculosis as an AIDS-qualifying clinical condition. This definition also adopted staging systems for HIV disease that distinguish three stages of CD4 depletion: 500/ml or above, 200-499/ml, and below 200/ml. This proposal permits staging using clinical data, where the patient is staged in one of three mutually exclusive stages. In this staging system, tuberculosis is considered as an AIDS-qualifying illness. In the revised staging system, CDC has mentioned that the inclusion of pulmonary tuberculosis as an AIDS-qualifying condition was " based on the strong epidemiologic link between HIV infection and the development of tuberculosis", and that "persons co-infected with HIV and tuberculosis have a substantially increased risk of developing active tuberculosis compared with persons without HIV infection". Another reason cited by CDC was that the addition of pulmonary tuberculosis was expected to have a substantial impact on the number of AIDS cases included in AIDS surveillance.

In Africa, by contrast, tuberculosis develops among HIV-infected persons across a broad range of CD4 counts, and is not necessarily linked to a specific stage of HIV disease and may not predict immunosuppression level in HIV-infected patients. In a study conducted in Kinshasa, Mukadi *et al.* observed that tuberculosis cases were

distributed nearly equally across the different stages of HIV disease [135]. Although patients infected with HIV and *Mycobacterium tuberculosis* were at increased risk of developing active tuberculosis, and this risk increases with increasing CD4 depletion or immunosuppression, the diagnosis of tuberculosis alone was not helpful in the staging of HIV disease.

Thus, it may be reasonable to regard tuberculosis as an AIDS-defining condition in developed countries, as proposed by the CDC, but not in Africa where tuberculosis is endemic. Tuberculosis in Africa, or any other setting where tuberculosis is prevalent, may not be associated with reduced survival as the case in the developed countries.

An ideal staging system for HIV should be universally applicable, reflect the pathophysiology of the disease, and simple to apply. A staging system for HIV should ideally have several important characteristics. Its mutually exclusive categories should reflect the entire range of clinical outcomes, from asymptomatic infection to serious disease, be clinically relevant and reflect increasing severity of illness, should include inexpensive and available laboratory tests, and should be associated with prognosis. The WHO stated that such a system could aid the study of the natural history of the disease, establish reliable prognoses, improve the clinical care of infected people, and provide a basis for the stratification of patients in vaccine and drug trials. Different stages could be used to provide standarized criteria to evaluate HIV disease progression in efficacy trials of therapeutic drugs or vaccines.

A number of studies have been undertaken to test the utility of the proposed staging systems [139,140,141,142]. The following limitations of the CDC staging system and

the studies on which this staging system and its successive revisions was based (in addition to the ones cited above) can be argued:

1. The CDC expanded definition requires laboratory confirmation of HIV infection in persons by applying CD4+ count techniques, which are impractical due to their unavailability in poor countries.

2. The follow-up period of patients in these studies was relatively short. Significant differences in the risk of progression from HIV-seropositivity to AIDS, on the basis of clinical criteria alone, may be apparent only with prolonged observation.

3. The number of subjects was relatively small, and this may have limited the power of these studies to detect statistically significant differences between the first stages and the last one which, according to the CDC staging system, included pulmonary tuberculosis as an AIDS defining condition.

4. The possibility of some misclassification bias cannot be excluded, because many of these studies were not designed specifically for the purpose of validating this staging system. The criteria for AIDS-staging may not have represented common manifestations of the disease in this population or any other population, especially those in African or other developing countries.

5. There remains the possibility of underreporting of the clinical endpoint, AIDS. Although these studies used several methods to identify their cohorts members

who had progressed to AIDS, disease may have not been reported because of reporting delays, emigration from the study region, or incorrect cause of death recorded on the death certificate.

6. Most of these studies were based on cohorts with predominantly either homosexual men or intra-venous drug users. Furthermore, such drug users present unique considerations in the staging of HIV infection and AIDS. Clinical signs such as lymphadenopathy and fever have been reported to be less specific for HIV infection among intra-venous drug users than in other high risk groups, because of the frequent association of such signs and symptoms with drug use itself. Survival of these patients may be confounded by the effect of drug use, which, in general, may explain the shorter survival of this subgroup [139]. This again leads one to question the generalisability of the findings of these studies to African or any other developing countries patients, where risk behaviours are different.

7. The recent changes in the CDC system, which expanded the AIDS surveillance case definition and increased the number of AIDS-defining illnesses, particularly tuberculosis, may further limit its utility as a universal staging system. Initially, this system was designed for public health surveillance and did not predict progression from HIV seropositivity to AIDS by category. More important, international acceptance of the revised system has been limited because of concerns that some of the changes, particularly the diagnosis of AIDS on the basis of laboratory criteria alone, may not reflect the pathophysiology of the disease. Because the epidemiology of HIV infection in

the developed countries is often markedly different from that of developing countries, this staging system, which defines tuberculosis as an AIDS-defining condition, must be evaluated among populations with different disease profiles or behavioural risks, *viz.* heterosexual, non- risk behaviour patients.

Very few studies have been done in Africa to evaluate HIV staging systems. Lifson *et al.* carried out a study in Kigali, Rwanda to evaluate the WHO staging system in 412 HIV-infected women recruited from prenatal and paediatric clinics [143]. On the basis of this evaluation, they proposed an HIV staging system for sub-Saharan Africa which included pulmonary tuberculosis as an AIDS-defining illness. They argued that "although tuberculosis may develop at an earlier stage of immune suppression, it may also be associated with a worse survival once it occurs" and that, according to previous data from their cohort, "5 (42%) of 12 HIV-infected women with pulmonary tuberculosis and no known extrapulmonary disease died within 15 months". They further argued that "in addition to death directly caused by tuberculosis, progression of HIV disease may be accelerated if tuberculosis adversely affects the immune system or enhances HIV expression. If so, this accelerated progression may lead to an increased risk for death from other immuno-deficiency-associated illnesses". However, this study suffers from the following limitations:

1. The study was exclusively based on a cohort of women recruited from prenatal and paediatric clinics. It has been observed that pregnancy induces immune depression in HIV-infected women pregnant women [19]. The reduced survival observed in this group might have been influenced by the immune depression which might have been experienced by these patients, and which

may confound the association between the excess mortality and pulmonary tuberculosis.

2. The shorter survival observed in this cohort was based on a very small sample; twelve patients only.

3. HIV-patients with active tuberculosis respond well to antituberculosis therapy, and usually experience an improved survival [55,61]. The authors of this study have not provided information regarding use of or compliance with antituberculosis therapy. It is difficult to exclude these factors as an alternative explanation for short survival observed in this study.

In general, the HIV-staging system proposed in the Lifson study, which defined pulmonary tuberculosis disease as an AIDS-defining condition, might have been based on patients with pulmonary tuberculosis who experienced poor survival for reasons other that pulmonary tuberculosis itself. Since factors such as advanced immune suppression were not controlled for in this study, it is difficult to disentangle their effect from the effect of pulmonary tuberculosis on survival. Thus, it is difficult to generalize the results of this study to other patients from sub-Saharan African countries.

In conclusion, where to place active tuberculosis disease in the HIV-staging system, particularly in African patients, remains a question of interest, since the answer to this question may have serious implications for the management and care delivered to patient in the African setting.

2-13 SITE OF DISEASE IN HIV-ASSOCIATED TUBERCULOSIS AND SURVIVAL

In previous studies of tuberculosis in HIV infected subjects, several predictors of poorer survival have been identified. These predictors included multiple drug resistance [144-145] and depressed immune status as measured by CD4+ count [43,55,146].

In addition to the above predictors of survival in HIV–infected patients with active tuberculosis, the site of tuberculosis disease has been shown to be an important prognostic factor. Shafer *et al.* have reported poor survival related to extrapulmonary tuberculosis [147]. Whalen *et al.* have shown that the site of extrapulmonary disease is an important prognostic factor, especially when the disease involves the meninges, blood, or bone marrow [136]. Among all HIV-infected patients with tuberculosis in their study, about 10% were diagnosed with tuberculous meningitis, which was associated with a poor prognosis. The mortality rate was high (44%) and the relative risk for death from tuberculous meningitis was high (relative risk = 3.9) compared with patients with pulmonary involvement alone.

Although some studies have thus reported poor survival related to extrapulmonary disease, this has not been evaluated in the context of other predictors of survival. There exists a need to ascertain whether (and which) site of tuberculous disease is associated with poorer survival in HIV-infected patients, particularly in TB endemic areas.

REFERENCES

1. Wood R, Maartens G, O'Keefe E. **The changing pattern of transmission and clinical presentation of HIV infection in the Western Cape region of South Africa (1984-1995).** *S Afr J Epidemiol Infect* 1996; 11 (4): 96-98.
2. Post F, Wood R. **HIV infection is not associated with an increased rate of drug-resistant tuberculosis.** *S Afr Med J* 1997; 87: 903.
3. Lundgren JD, Pedersen C, Clumeck N, et al. **Survival differences in patients with AIDS, 1979-1989.** *Br Med J* 1994; 308: 1068-1073.
4. Rothenberg R, Woelfel M, Stoneburner R, Milberg J, Parker R, Truman B. **Survival with the acquired immunodeficiency syndrome - experience with 5833 cases in New York city.** *N Engl Med* 1987; 317: 1297-1302.
5. Lemp GF, Payne SF, Neal D, Temelso T, Rutherford GW. **Survival trends for patients with AIDS.** *JAMA* 1990; 263: 402-406.
6. Whitmore-Overton SE, Tillet HE, Evans BG, Allardice GM. **Improved Survival from diagnosis of AIDS in adult cases in the United Kingdom and bias due to reporting delays.** *AIDS* 1993; 7: 415-420.
7. Chang HH, Morse DL, Noonan C, et al. **Survival and mortality patterns of an acquired immunodeficiency syndrome (AIDS) cohort in New York state.** *Am J Epidemiol* 1993; 138: 341-349.
8. Lou K, Law M, Kaldor JM, McDonald AN, Cooper DA. **The role of initial AIDS-defining illness in survival following AIDS.** *AIDS* 1995; 9: 57-63.
9. Jacobson LP, Kirby AJ, Polk S, et al. **Changes in survival after acquired immunodeficiency syndrome (AIDS): 1984-1991.** *Am J Epidemiol* 1993; 138:952-964.

10. Stehr-Green JK, Holman RC, Mahoney MA: **Survival analysis of hemophilia-associated AIDS cases in the USA.** *Am J Pub Health* 1989; 79: 832-835.

11. Reeves GK, Overton SE. **Preliminary survival analysis of UK AIDS data [letter],** *AIDS* 1988; i: 880.

12. Seage GR III, Oddleifson S, Carr E, et al. **Survival with AIDS in Massachusetts 1979-1989.** *Am J Pub Health* 1993; 83: 72-78.

13. Bacchetti P, Osmond D, Chaisson RE, et al. **Survival patterns of the first 500 patients with AIDS in San Francisco.** *J Infect Dis* 1988; 157: 1044-1047.

14. Marasca G, McEvey M. **Length of survival of patients with acquired immune deficiency syndrome in the United Kingdom.** *Br Med J* 1986; 292: 1727-1729.

15. Chaisson RE, Keruly JC, Moore RD. **Race, sex, drug use, and progression of human immunodeficiency virus disease.** *N Engl J Med* 1995; 333: 75 1-756.

16. Philips AN, Elford J, Sabin C, Bofill M, Janossy G, Lee CA: **Immunodeficiency and the risk of death in HIV infection.** *JAMA* 1992; 268: 2662-2666.

17. Colford JM Jr, Ngo L, Taer I. **Factors associated with survival in human immunodeficiency virus infected patients with very low CD4 counts.** *Am J Epidemiol* 1994; 193: 206-218.

18. Martin DJ, Sim JG, Sole GJ, et al. **CD4+ T-lymphocyte count in African patients co-infected with HIV and tuberculosis.** *J Acquir Imuune Defic Syndr Hum Retrovirol* 1995; 8: 386-391.

19. Rich KC, Siegel JN, Jennings C, et al. **CD4+lymphocytes in peri-natal human immunodeficiency virus (HIV) infection: evidence for pregnancy induced immune depression in uninfected and HIV-infected women.** *J Infect Dis* 1995; 172: 1221-1227.

20. Hogg RS, Strathdee SA, Craib KJP, Shaugnessy MVO, Montaner JSG, Schechter MT. **Lower socioeconomic status and shorter survival following HIV infection.** *Lancet,* 1994; 344: 1120-1124.

21. Apolonio EG, Hoover DR, He Y, et al. **Prognostic factors in human immunodeficiency virus-positive patients with CD4+ lymphocyte count <50/l.** *J Infect Dis* 1995; 171: 829-836.

22. Eskild A, Magnus P, Nilsen O, Hasseltvedt V, Lystad A. **Asymptomatic subjects at HIV diagnosis have prolonged survival as AIDS patients.** *Int J Epidemiol* 1992; 21: 387-390.

23. Pozansky MC, Coker R, Skinner C, et al. **HIV positive patients first presenting with an AIDS defining illness: characteristics and survival.** *Br Med J* 1995; 311: 156-158.

24. Reed GM, Kemeny ME, Taylor SE, Wang HY, Visscher BR. **Realistic acceptance as a predictor of decreased survival time in gay men with AIDS.** *Health Psych* 1994; 13: 299-307.

25. Laraque F, Greene A, Triano-Davis JW, et al. **Effect of comprehensive intervention program on survival of patients with human immunodeficiency virus infection.** *Arch Intern Med* 1996; 156: 169-179.

26. Diamondstone LS, Blakely SA, Rice JC, et al. **Prognostic factors for all cause mortality among hemophiliacs infected with human immunodeficiency.** *Am J Epidemiol* 1995; 142: 304-313.

27. Scharschmidt BF, Held MJ, Hollander HH, et al. **Hepatitis B in patients with HIV infection : relationship to AIDS and patient survival.** *Ann Intern Med* 1992; 117: 837-838.

28. Tovo PA, de Martino M, Gabiano C, et al. **Prognostic factors and survival in children with prenatal HIV-1 infection**. *Lancet* 1992; 339: 1249-1253.

29. Drew WL, Miner RC, et al. **Prevalence of cytomegalovirus infection in homosexual men** . *J Infect Dis* 1981; 143: 188-192.

30. Mintz L, Drew WL, Miner RC, Braff EH. **Cytomegalovirus infection in homosexual men : an epidemiological study**. *Ann Intern Med,* 1983; 98: 326-329.

31. Sabin CA, Philips AN, Lee CA, Janossy G, Emery V, Griffiths PD: **The effect of CMV infection on progression of human immunodeficiency virus diseases in a cohort of hemophiliac men followed for up to 13 years from seroconversion.** *Epidemiol Infect* 1995; 114: 361-372.

32. Lundgren JD, Melbye M, Pederson C et al. **Changing patterns of Kaposi's sarcoma in Danish acquired immunodeficiency syndrome patients with complete follow-up**. *Am J Epidemiol* 1995; 141: 652-658.

33. Bulterys M, Nzabihimana E, Chao A et al: **Long term survival among HIV-1-infected prostitutes** [Letter]. *AIDS* 1993; 7: 1269.

34. Veugelers PJ, Page KA, Tindall B, et al. **Determinants of HIV disease progression among homosexual men registered in the Tri-continental converter study**. *Am J Epidemiol* 1994; 140:7 47-758.

35. Lykestsos CG, Hoover DR, Guccione M et al. **Depressive symptoms as predictors of medical outcomes in HIV infection**. *JAMA* 1993; 270:2563-2567.

36. Blum S, Singh TP, Gibbons J et al. **Trends in survival among persons with acquired immunodeficiency syndrome in New York City. The experience of the first decade of the epidemic**. *Am J Epidemiol* 1994; 139: 351-361.

37. Centres for Disease Control. **1993 revised classification system for HIV infection and expanded surveillance case definition for AIDS among adolescents and adults.** *MMWR,* 1992; 11:1-9

38. Johnson AM, De Cock KM. **What is happening to AIDS?** *Br Med J,* 1994; 309: 1523-1524.

39. Kochi A. **The global tuberculosis situation and the new control strategy of the World Health Organization**. *Tubercle Lung Dis* 1992; 72: 1-6.

40. Grange JM. **Mycobacteria and Human Disease.** *Oxford Press* 1996.

41. Dolin PJ, Raviglione MC and Kochi A. **Global tuberculosis incidence and mortality during 1990-2000.** *Bullet World Health Org* 1992; 72: 213-20.

42. Sundre P, Dan GT, Kochi A. **Tuberculosis: a global overview of the situation today**. *Bullet World Health Org* 1992; 70: 149-159.

43. Colebunders RL, Ryder RW, Nzilanbi N, et al. **HIV infection in patients with tuberculosis in Kinshasa, Zaire**. *Am Rev Respir Dis* 1989; 139: 1082-1085.

44. Narain JP, Raviglione MC and Kochi M. **HIV-associated tuberculosis in developing countries : epidemiology and strategies for prevention.** *Tubercle Lung Dis* 1992; 73: 311-21.

45. Braun MM, Badi N, Ryder RW et al. **A retrospective cohort study of the risk of tuberculosis among women of childbearing age with HIV infection in Zaire**. *Am Rev Respir Dis* 1991; 143: 501-504.

46. Bouvet E, Casalino E, Mendoza-Sassi G et al. **A nosomical outbreak of multidrug-resistant *Mycobacterium bovis* among HIV-infected patients. A case-control study**. *AIDS* 1993; 7:1453-60.

47. Schulzer M, Fitzgerald JM, Enarson DA and Grzybowski S. **An estimate of the future size of the tuberculosis problem in sub-Saharan Africa resulting from HIV infection.** *Tubercle Lung Dis* 1992; 73: 52-8.

48. Rieder HL, Canthen GM, Kelly GD, Bloch AB, Snider DE. **Tuberculosis in the United States.** *JAMA* 1989; 262: 385-9.

49. **Tuberculosis - United States, 1985-and possible impact of the Human Lymphotropic Virus type III/Lymphadenopathy-associated virus infection.** *MMWR* 1986; 35: 74-6.

50. Triol I, Schwoebel V, Delmas MC, Pidget R, Laporte A, Brunet J-B. **Factors associated with tuberculosis at AIDS diagnosis in France.** *AIDS* 1996; 10: 223-228.

51. Elliot A, Luo N, Tempo G, et al. **The impact of human immunodeficiency virus in Zambia : A cross-sectional study.** *Br Med J* 1990; 301:412.

52. Nunn P, Kibunga D, Elliot A, **The impact of human immunodeficiency virus on transmission and severity of tuberculosis.** *Trans R Soc Trop Med Hyg* 1990; 84 (suppl. 1:9-13)

53. Chaisson R, Schecter G, Theuer C, Rutherford G, Echenberg D, Hopewell P. **Tuberculosis in patients with acquired immunodeficiency syndrome.** *Am Rev Respir Dis* 1987; 136: 570-4.

54. Mukadi y, Perriens J, Willame J.C, et al. **Short course antituberculosis therapy for pulmonary tuberculosis in HIV positive patients : A Prospective controlled study.** *N Engl J Med* 1990; 248.

55. Nunn P, Bridle R, Carpenter L et al. **Cohort study of human immunodeficiency virus infection in patients with Tuberculosis in Nairobi, Kenya.** *Am Rev Respir Dis* 1992; 148: 899-854.

56. Eriki PP, Okwera A, Aisu T et al. **The influence of human immunodeficiency virus infection on tuberculosis in Kampala, Uganda**. *Am Rev Respir Dis* 1991; 143: 185-7.

57. De Cock KM, Gnaore E, Adjorlolo G et al. **Risk of tuberculosis in patients with HIV-1 and HIV-II infections in Abidjan, Ivory Coast**. *BMJ* 1991; 302: 496-499.

58. Wood R, Post FA. **Survival of human immunodeficiency HIV virus-infected persons with pulmonary tuberculosis**. *Int J Tuber Lung Dis* (1): 87.

59. Slavik T, Wolfaardt M, Van Zyl H, Simon IW. **HIV-1 infection for patients with tuberculosis Lymphadenitis**. *S Afr Med J* 1996; 86: (1) 92.

60. Selwyn PA, Hartel D, Lewis VA et al. **A prospective study of the risk of tuberculosis among intravenous drug users with human immunodeficiency virus infection**. *N Eng J Med* 320; 545-550.

61. Chaisson R, Schecter G, Theuer CP, Rutherford GW, Echenberg DF, Hopewell PC. **Tuberculosis in patients with the acquired immunodeficiency syndrome-Clinical features, response to therapy and survival.**. *Am Rev Respir Dis* 1987; 136: 570-574.

62. Wilkinson D, Moore DAJ. **HIV-related tuberculosis in South Africa-clinical features and outcomes**. *S Afr Med J* 1996; 86: 60-63.

63. Department of National Health and Population Development. **Seventh national HIV survey of women attending antenatal clinics. South Africa**. *Epidemiol Comments* 1996; 23(2): 4-16.

64. Doyle PR. **The impact of AIDS on the South Africa population. AIDS in South Africa: the Demographic and Economic Implications**. *Johannesburg: Centre for Health Policy, University of the Witwatersand; 1991.*

65. Lee T, Esterhuyse T, Steinberg M. **Demographic Modelling of the HIV/AIDS Epidemic on Sweto Population**. *Johannesberg: Centre for Health Policy, University of the Wirwatersand 1993.*

66. Beyers N, Gie RP, Zietsman HL, et al. **The use of Geographical information system (GIS) to evaluate the distribution of tuberculosis in a high incidence community**. *S Afr Med J* 1996; 86: 40-44.

67. Department of National Health and population Development, republic of South Africa. **Exploring some hypothesis on tuberculosis with a distinguished visitor from abroad**. *Epidemiol Com* 1993; 20:90-100.

68. Desrosiers RC. **HIV-1 origins. A finger on the missing link**. *Nature* 1990; 345:288-289.

69. Rosenberg ZF, Fauci AS. **The immunopathogenesis of HIV infection**. *Adv Immunol* 1989; 47: 377-431

70. Desrosiers RC, Hansen-Moosa K, Moori DP, et al. **Macrophages-tropic variants of SIV are associated with specific AIDS-related lesions but are not essential for development of AIDS**. *Am J Pathol* 1991; 139:29-35.

71. Vicezi E, Biswas P, Mengozzi M, Poli G. **Role of pro-inflammatory cytokines and beta-chemokines in controlling HIV replication**. *J Leukoc Biol* 1997; 62: 34-40.

72. Dalgleish AG, Beverly Pr, Clapham DH, Crawford, Greaves MF, Weiss RA. **The CD4(4) antigens is an essential component of the receptor for the AIDS retrovitus**. *Nature* 1984; 312: 763-767.

73. Orenstien JM, Meltzer MS, Phipps T, Gendelman HE. **Cytoplasmic assembly and accumulation of human immunodeficiency virus types 1 and 2 in**

recombinant human colony-stimulating factor-1-treated human monocytes: An ultrastructureal study. *J Virol* 1988; 62: 2578-2586.

74. McDougal JS, Mawle SP, Cort JKA, et al. **Cellular tropism of the human retrovirus HTLV-III/LAV.I. Role of T cell activation and expression of the T4 antigen.** *J Immunol* 1985; 135:3151-3162.

75. Emberson J, Zupanic JL, Ribas WT, et al. **Massive covert infection of helper T lymphocytes and macrophages by HIV during the incubation period of AIDS.** *Nature* 1993; 362: 359,362.

76. Straub OC. **Caprine arthritis encephalitis – A model for AIDS.** *Intervirol* 1989; 30:45-50.

77. Smith PD, Quinn TC, Strober QW, Janof EN, Masur H. **NIH conference: Gastrointestinal infections in AIDS.** *Ann Intern Med* 1992; 116:63-77.

78. Fauci AS. **The human immunodeficiency virus: infectivity and mechanisms of pathogenesis.** *Science* 1988; 239: 617-622.

79. Poli G, Fauci AS. **The effects of cytokines and pharmacologic agents on chronic HIV infection.** *AIDS Res Hum Retroviruses* 1992; 8: 191-197.

80. Yamamoto N. **The role of cytokines in the acquired immunodeficiency syndrome.** *Int J Clin Lab Res* 1995; 25: 29-34.

81. Pantaleo G, Graziosi C, Fauci AS. **The immunopathogenesis of Human Immunodeficiency Virus Infection.** *N Engl Med* 1993; 328: 327-335.

82. Tindall B, Cooper DA. **Primary HIV infection: host responses and intervention strategies.** *AIDS* 1991; 5: 1-14.

83. Piatak M, Saag MS, Yang LC, et al. **High levels of HIV-1 in plasma during all stages of infection determined by competitive PCR.** *Science* 1993; 259: 1749-1754.

84. Koup RA, Safrit JT, Cao Y et al. **Temporal associations of cellular immune responses with the initial control of viremia in primary human immunodeficiency virus type 1 syndrome.** *J Virol* 1994; 68:4650-4655.

85. Moss AR, Bacchetti. **Natural history of HIV infection.** *AIDS* 1989; 3: 55-61.

86. Lemp GF, Payne SF, Rutherford GW, et al. **Projections of AIDS morbidity and mortality in San Francisco.** *J Am Med Assoc* 1990; 263: 1497-1501.

87. Perelson AS, Essunger P, Cao Y, et al. **Decay characteristics of HIV-1-infected compartments during combination therapy.** *Nature* 1997 397: 188-191.

88. Allen BW, Hinkes WF. **Koch's stain for tubercle bacilli.** *Bullet Int Union Tuberc* 1982; 57: 190-192.

89. Abou-Zaid C, Smith I, Grange JM, et al. **The secreted antigens of *mycobacterium tuberculosis* and their relationship to those recognized by the available antibodies.** *J Gen Microbiol* 1988; 134: 531-535.

90. Critchley M. ***Butterworth Medical Dictionary.*** 1978; *London; Butterworths.*

91. Roth J, Bolin C, Brogden K, Minion F, Wannemuehler J. **Virulence mechanism of bacterial pathogens.** 1995; *Washington DC; American Society of Microbiology.*

92. Bloom BR. **Revisiting and revising suppressor T cells.** *Immunol Today* 1992; 13:131-136.

93. Dannenberg AM. **Immunopathogenesis of pulmonary tuberculosis.** *Hosp Practice* 1993; 28: 51-58.

94. Lorgat F, Keraan MM, Lukey PT, Ress SR. **Evidence for *in vivo* generation of cytotoxic T cells: PPD-stimulated lymphocytes from tuberculosis pleural effusions demonstrate enhanced cytotoxicity with accelerated kinetics of induction.** *Am Rev Resir Dis* 1992; 145: 418-423.

95. Mosmann TR, Moore KW. **The role of IL-10 in crossregulation of TH1 and TH2 responses.** *Immunol Today* 1991;12: 49-53.

96. Ainsile GM, Solomon JA, Bateman ED. **Lymphocyte and lymphocyte subsets numbers in blood and bronchoalveolar lavage and pleural fluid in various forms of human pulmonary tuberculosis at presentation and during recovery.** *Thorax* 1992; 47: 513-518.

97. Brostoff JL, Lenzini L, Rottoli P, Rottoli L. **Immune response in the spectrum of tuberculosis.** *Tubercle* 1981 62;169-173.

98. Flynn JL, Goldstien MM, Triebold KL, et al. **Major histocompatibility complex calss I-restricted T cells are required for resistence to Mycobacterium tuberculosis infection.** *Proceedings of the National Academy of science of the USA* 1991; 89: 12013-7.

99. Beck JS, Morley SM, Lowe JG, et al. **Diversity in migration of CD4 and CD4 lymphocytes in different microanatomical compartments of the skin in tuberculin reaction in man.** *Br J Exper Pathol* 1988; 60: 49-59.

100. Rook GAW. **The role of vitamin D in tuberculosis.** *Am Rev Respir Dis* 1986;138: 768-770.

101. Flesch I, Kaufmann S. **Activation of tuberculostatic macrophage functions by gamma interferon, interleukin-4 and tumor necrosis factor.** *Infect Immunol* 1990; 58:2675-2677.

102. Beutler B, Greenwald D, Hulmes JD, et al. **Identity of tumour necrosis factor and the macrophage-secreted factor cachectin.** *Nature* 1985; 316:552-553.

103. Orme IM, Anderson P, Boom WH. **T cell response to *Mycobacterium tuberculosis*.** *J Infect Dis* 1993; 167:1481-1497.

104. McDonough KA, Kress Y, Bloom BR. Pathogenesis of tuberculosis: interactions of ***Mycobacterium tuberculosis*** **with macrophages.** *Infect Immunol* 1993; 61: 2763-2673.

105. Filley EA, Bull HA, Dowd PM, Rook GAW. **The effect of to Mycobacterium tuberculosis on the susceptibility of human cells to the stimulatory and toxic effects of tumour necrosis factor.** *Immunol* 1992; 77: 505-509.

106. Gibbs JH, Grange JM, Beck JS, et al. **Early delayed-type hypersensitivity responses in tuberculin skin tests after heavy occupational exposure to tuberculosis.** *J Clin Pathol* 1991; 44: 919-923.

107. Wallis RS, Vejecha M, Amir-Tahmasseb N, et al. **Influence of tuberculosis on human immunodeficiency virus (HIV-1): enhanced cytokine expression and elevated β_2-microglobulin in HIV-1-assocciated tuberculosis**. *J Infect Dis* 1993; 167:43-48.

108. Garrait V, Cadranel J, Esvant H, et al. **Tuberculosis generates a microenvironment enhancing the productive infection of local lymphocytes by HIV.** *J Immunol* 1997; 159: 2824-2830.

109. Goletti D, Weissman D, Jackson RW. **Effect of *Mycobacterium tuberculosis* on HIV replication: role of immune activation**. *J Immunol* 1996; 157: 1271-1278.

110. Zhang Y, Nakata K, Weiden M, Rom WN. ***Mycobacterium tuberculosis*** **enhances human immunodeficiency virus-1 replication by transcriptional activation at the long terminal repeat.** *J Clin Invest* 1995; 95; 2324.

111. Zhou D, Shen Y, Chalifoux L, et al. ***Mycobacterium bovis*** **Bacille Calmette-Guerin enhances pathogenicity of simian immunodeficiency virus infection and accelerates progression to AIDS in macaques: A role of persistent T cell activation in AIDS pathogenesis.** *J Immunol* 1999; **162:2204-2216.**

112. Ellner JJ. **Tuberculosis in the time of AIDS.** *Chest* **1990; 98: 1051.**

113. Pitchenik AE, Cole C, Russell BW, Fischl MA, Spira TJ, Snider DE. **Tuberculosis, atypical *mycobacteriosis*, and the acquired immunodeficiency syndrome among Haitian and non-Haitian patients in South Florida.** *Ann Intern Med* 1984; 101: 641-5.

114. Sundram G, McDonald RJ, Maniatis T, Oleske J, Kapila R, Reichman LB. **Tuberculosis as a manifestation of the acquired immunodeficiency syndrome (AIDS).** *JAMA* 1986; 256: 362-6.

115. Modilevsky T, Sattler FR, Barnes PF. **Mycobacterial disease in patients with HIV infection** . *Arch Intern Med* 1989; 149: 2201-5.

116. Kramer F, Modilevsky T, Waliany AR, Leedom JM, Barnes PF. **Delayed diagnosis of tuberculosis in patients with human immunodeficiency virus infection.** *Am J Med* 1990; 89: 451-6.

117. Kelly P, Burnham G, Radford et al. **HIV seropositivity and tuberculosis in rural Malawi hospital.** *Trans Roy Soc Trop Med Hyg* 1990; 84:725-727.

118. Bethlem N. **AIDS and tuberculosis in Brazil.** *AIDS* 1989; 50: 19-21.

119. Pitchenik AE, Robinson HA. **The radiographic appearance of tuberculosis in patients with acquired immunodeficiency syndrome (AIDS) and pre-AIDS.** *Am Rev Respir Dis* 1985;131: 393-396.

120. Harries AD. **Tuberculosis and human immunodeficiency virus infection in developing countries.** *Lancet* 1990; 335: 387-390.

121. Long R, Scalcini, Manfreda J, et al. **Impact of human immunodeficiency virus type 1 on tuberculosis in rural Haiti.** *Am Rev Respir Dis* 1991; 143: 69-73.

122. Tshibwabwa-Tumba E, Mwinga A, Pobee JO, Zumla A. **Radiological features of pulmonary tuberculosis in 963 HIV-infected adults at three Central African Hospitals.** *Clin Radiol* 1997; 52 (11): 837-41.

123. Small PM. **Treatment of tuberculosis in patients with advanced human immunodeficiency virus infection.** *N Engl J Med* 1991; 324: 289-294.

124. Gilks CF, Brindle RJ, Otieno LS et al. **Extrapulmonary and disseminated tuberculosis in HIV-seropositive patients presenting to the acute medical services in Nairobi.** *AIDS* 1990; 4: 981-985.

125. Horsburgh CR, Pozniak A. **Epidemiology of tuberculosis in the era of HIV.** *AIDS* 1993;7(suppl 1): S 109-114.

126. Chequer P, Hearst N, Hudes ES, Castilo E, Rutherford G, Loures L. **Determinants of survival in adult Brazilian AIDS paients, 1982-1989.** *AIDS* 1992; 6: 483-487.

127. Perneger TV, Sudre P, Lundgren JD, Hirschel B. **Does the onset of tuberculosis in AIDS predict shorter survival** ? *Br Med J* 1995; 311: 1468-1471.

128. Buira E, Gatell JM, Miro JM, Batalla J, Zamora L, Mallolas J. **Influence of treatment with Zidovudine (ZDV) on the long-term survival of AIDS patients.** *AIDS* 1992; 5: 737-742.

129. Lundgren JD, Pedersen C, Clumeck N, Gatell JM, Johnson AM, Ledergerber B. **Survival differences in European patients with AIDS, 1979-89.** *Br Med J* 1994; 308: 1068-1073.

130. Whalen C, Horsburgh RC, Hom D, Lahart C, Simberkoff M, Ellner J. **Accelerated course of human immunodeficiency virus infection after tuberculosis.** *Am J Respir Crit Care Med* 1995; 151: 129-135.

131. Leroy V, Salmi R, Dupon M, etal. **Progression of human immunodeficiency virus in patients with tuberculosis disease.** *Am J Epidemiol* 1997; 145: 293-300.

132. Stoneburner R, Laroche E, Prevots R. **Survival in a cohort of human immunodeficiency virus-infected tuberculosis patients in New York City.** *Arch Intern Med* 1992; 152:2033-2037.

133. Murray CJ, Styblo K, Rouillon A. **Tuberculosis in developing countries: burden, intervention and cost.** *Tubercle Lung Dis* 1990; 65: 6-24.

134. Munsiff SS, Alpert PL, Gourevitch MN, chang CJ, Klein RS. **A prospective study of tuberculosis and HIV disease progression.** *J Acquir Immune Defic Syndr Hum Retrovirol* 1998; 19: 361-366.

135. Mukadi Y, perriens JH, Louis ME, et al. **Spectrum of immunodeficiency in HIV infected patients with pulmonary tuberculosis in Zaire.** *Lancet* 1993; 342: 143-146.

136. Whalen C, Horsburgh RC, Hom D, Lahart C, Simberkoff M, Ellner J. **Site of disease and opportunistic infection predict survival in HIV-associated tuberculosis.** *AIDS* 1997; 11: 455-460.

137. Pitchenik AE. **Tuberculosis control and the AIDS epidemic in developing countries.** *Ann Intern Med* 1990; 133: 89-91.

138. Mukadi Y, perriens JH, Louis ME, et al. **Spectrum of immunodeficiency in HIV infected patients with pulmonary tuberculosis in Zaire.** *Lancet* 1993; 342: 143-146.

139. Selwyn PA, Hartel D, Lewis VA, et al. **A prospective study of the risk of tuberculosis among intravenous drug users with Human immunodeficiency virus infection.** *N Engl J Med* 1989; 320: 545-550.

140. Buehler JW, Devine OJ, Berkelman RL, Chevarley FM. **Impact of the human immunodeficiency virus epidemic on mortality trends in young men, United States.** *Am J Pub Health* 1990; 80: 1080-1086.

141. Montaner JSG, Le TN, Craib KJP, Schecter MT. **Application of the WHO system for HIV infection in a cohort of homosexual men in developing a prognostically meaningful staging system.** *AIDS* 1992; 6: 719-724.

142. Aylward BR, Vlahov D, Munoz A, Rapiti E. **Validation of the proposed World Health Organization staging system for HIV disease and infection in a cohort of intravenous drug user.** *AIDS* 1994; 8: 1129-1133.

143. Lifson AR, Allens, Wolfson W, et al. **Classification of HIV infection and disease in women from Rwanda. Evaluation of the World Health Organisation HIV-staging system and recommended modifications.** *Ann Intern Med* 1995; 22: 262-270.

144. Shafer RW, Small PM, Larkin C, et al. **Temporal trends and transmission patterns during the emergence of multidrug-resistant tuberculosis in New York City : a molecular epidemiologic assessment.** *J Infect Dis* 1995; 171:170-176.

145. Fischl MA, Daikos GL, Uttamchandani RB, et al. **Clinical presentation and outcome of patients with HIV infection and tuberculosis caused by multi-drug-resistant bacilli.** *Ann Intern Med* 1992; 117: 184-190.

146. Whalen C, Okwera A, Johnson J, et al. **Prognosis and survival in HIV-infected individuals with tuberculosis : a comparison of US and Ugandan cohorts** [abstract]. *Tubercle Lung Dis* 1994; 75 (suppl 1): 66A.

147. Shafer RW, Kim DS, Weiss JP, Quale JM. **Extrapulmonary tuberculosis in patients with human immunodeficiency virus infection.** *Medicine* 1991; 70: 384-397.

3. METHODS

3.1 STUDY DESIGN

Prospective patient cohort study.

3.2 POPULATION AND SAMPLING

This study took place at the adult HIV clinics of University of Cape Town affiliated hospitals, New Somerset (NSH) and Groote Schuur (GSH). These clinics, which were established in 1984 (NSH) and 1992 (GSH), serve patients from across the sociodemographic spectrum of the Western Cape province. At that time, no other facilities in the public sector were available. All adult HIV seropositive patients presenting as an outpatient or inpatient from January 1992 to December 1996 were included in the study.

Inclusion criteria for patients were:

1. Confirmed HIV infection;
2. At least 18 years of age; and
3. Seen at one of the study sites on more than one occasion between 1992 and 1996.

To identify cases of pulmonary and extrapulmonary tuberculous disease among all patients in the cohort, the records completed by the clinicians during the follow-up were used. The tuberculosis case definition used was a positive culture or a compatible clinical picture combined with a positive smear or a histologic diagnosis.

Patients diagnosed with active tuberculosis were treated with 6-month short course regimen. Both antiretroviral and co-trimoxazole therapies were self-administered. Co-trimoxazole was prescribed for both TB cases and non-cases when CD4+ T lymphocyte count reached 200 cells/μL.

Patients were excluded from the study if:

1. Active tuberculosis had been diagnosed before 1992;
2. Tuberculosis diagnosis was not confirmed, and
3. They did not have a CD4+ lymphocyte count done within the 6 months before or after tuberculosis diagnosis.

HIV diagnosis was confirmed by ELISA and/or Western blot on 2 separate blood specimens by the Department of Virology, University of Cape Town.

Clinicians enrolled and followed the patients using a standard computerised format for recording epidemiologic, medical and biologic information. Follow-up was repeated at least every 6 months, and a repeated search for patients lost to follow-up was carried out to confirm their vital status. Demographic details, HIV risk behaviours, clinical diagnosis and follow-up visits were prospectively collected and entered into a computer database (Epi Info 6.01). The HIV clinic's records were the

primary source of information about sex, sociodemographic or ethnic group, date and place of birth, migration status, date of tuberculosis diagnosis, site of tuberculosis, results of mycobacteriologic evaluations, type and duration of antituberculosis therapy, status of tuberculosis at the most recent follow-up, and date of death.

At each attendance, patients were examined for HIV related manifestations and staged using the proposed WHO clinical HIV staging system (Appendix II). CD4 + T-cell counts were measured approximately 6 monthly, by flow cytometry at the South African Institute of Medical Research (SAIMR) laboratory, Cape Town or the Department of Immunology, University of Cape Town.

3.3 MEASUREMENTS

Using the computer database and chart review of patients at each clinic, it was possible to glean the sociodemographic information, CD4+ lymphocyte count, date of death, treatment for HIV infection, and occurrence of AIDS-defining illnesses, as defined by CDC classification for HIV infection [1]. CD4 + counts were checked for validity by comparing each measured CD4+ count with the preceding and succeeding count to ascertain that the CD4 at date of diagnosis value was consistent with the preceding and succeeding counts. Tuberculosis was further classified as pulmonary and extrapulmonary according to the radiologic, microbiologic and clinical examinations.

The list of patients dually infected with tuberculosis and HIV was cross-checked against the Provincial Tuberculosis Registry and were found in that database.

3.4 DEFINITION OF CASES AND NON-CASES (CONTROLS)

In this patient cohort study, the "exposure" is active tuberculosis. Patients who had received a confirmed diagnosis of active tuberculous disease (exposed = "cases") were compared with HIV-infected patients without tuberculous disease (unexposed = "controls"). All the patients seen at the two hospitals' HIV clinics between 1992 and 1996 with confirmed HIV positive status and who remained free of active tuberculosis until the end of the study were thus included in the study as controls.

The person-month total for each comparison group was defined as the time each patient contributed to the total person-months of his/her group from date of inclusion into the study to date of death or censoring, summed across the total number of patients in the group.

Comparisons between "cases" and "controls" included baseline characteristics (sociodemographic features and risk behaviour, history of AIDS-defining illnesses, treatment of HIV infection, and CD4+ lymphocyte count). The incidence density rate of AIDS-defining illnesses in the two groups was defined as the number of AIDS-defining events occurring in patients without an AIDS-defining illness at baseline per 100 person-years of follow-up. Follow-up of a patient was defined as the period from inclusion to December 1996, to the date of last visit before the study endpoint, or to death.

From: motasim badri <motasimb@hotmail.com>
To: GASNAT SHABOODIEN <gasnat.shaboodien@uct.ac.za>
Date: 2007/08/28 11:12 am
Subject: RE: Analysis

Dear Gasnat

Many thanks for the detailed message.

It is part of doing a PhD to get handle of the methods you are using. I know it is a great pain- I have been there before and it is frustrating. However, for more "practical" and "real-life" example, I suggest you look-up my MScMed thesis in the medical school library. It is entitled: "The prognosis of tuberculosis in adults infected with human immunodeficiency virus, university of Cape Town, South Africa" particularly from page 103 to a further few pages illustrating use of MH. Remember that Breslow-Day test is the main test there. The Qs addressed in the thesis is quite identical to yours. Just ask the libarian or do a search under "Badri".

To make things easier, I will re-write the analysis stuff that I sent you. You just need to re-produce it and you will be safe; if you decide that MH is the way to go, which I think is the appropriate analysis.

Will get to you later this day.

Best

MB> Date: Mon, 27 Aug 2007 10:19:21 +0200> From: gasnat.shaboodien@uct.ac.za> To: motasimb@hotmail.com> Subject: RE: Analysis> > Salaam Motasim,> let's finish the business section first ? .... :-)> We are looking into maybe using your Mantel-Haenszel analysis for my work. As there might be possible confounders in the groups it might be better to use the MH test. What I know about stats can be written on a pinhead, so to answer you questions I went online to try and find out what exactly Mantel-Haenszel was..... phew!.....I think I got the gist of it...I think the one website said that if confounders exist/or might exist MH would be the appropriate test to do in most study types, right? Well, you saw the groups we're dealing with as well as the questions we're asking...so maybe MH would be the better way to go?(OK, wait...I I've made a decision...please use MH, it seems more robust..). Bongani has also said that maybe we should look into using the MH data. I have a Q for you....there was no diff. in the OR, p-values, CI of the MH or Chir-square, so does it matter which one we use? > Hope you don't think I'm an idiot but would you mind listing the pros vs cons for using MH instead of Chi-sqaure? If you put down the issues in point form I can address each issue, because right now I don't know what the issues ar with either of the tests.I've just always used chi-square. > One more thing, I've tried to work out the power calculations for the study but have hit a brick wall...would you mind doing that for the two studies kanalla? Do we have sufficient power to detect an association if one existed?The (1) replicative study comprises the HIV-DCM+ (n=99) and the HIV-DCM- (n=117) groups and the (2) HIVAC study comprises the HIV+DCM+ (n=30), HIV+DCM- (n=38) and HIV-DCM- (n=117) groups. > > Phew, I think that's about it....hope I made some sense....if not, I'm sorry for confusing you!...> > As for Ramadaan?....everyone is gearing up for Ramadaan here in Cape Town...all you hear are people talking about what they're going to be eating, drinking, doing....it's really a very special time of the year here in CT...I wouldn't want to be anyhwere else!....am I making you miss CT???....:-)) well the days have definitely gone shorter for one...no more fasting 16-18 hours!....it's now just over 12 hours..> As for the weather?...for the past few years we only really started getting summer in November, so I'm expecting a cold Ramadaan with a few warm days (22 degrees) thrown in just for the heck of it....we're all still wearing boots, rain coats,scarves,etc and it doesn't look like we'll be packing these away any time soon.... hmmm nice, hot soup to go with all that deep fried 'baddies'....can't wait...> > What is Ramadaan like in the UK?> Keep well. > Salaam!> Gasna> > > >>> motasim badri <motasimb@hotmail.com> 2007/08/24 05:40 pm >>>> > Salam Gasnat> > In fact I was waiting for both, but as you suggest we can do them bit by bit. Will add my descriptive notes on the tables. > I am still have to hear from you about the Qs I sent you earlier alongwith these analysis. They will help me write the more > appropriate notes on them. I will answer your Qs in details after you answer mine. Please note Monday is a holiday, > but I promise to dedicate the whole of Tuesday for you. By the way, how do you expect Ramadan to be in RSA; hot or cold?> > > Fi aman ellah> MB> > > Date: Fri, 24 Aug 2007 13:20:12 +0200From: gasnat.shaboodien@uct.ac.zaTo: motasimb@hotmail.comSubject: Fwd: RE: AnalysisSalaam

Patients were considered "free of tuberculosis" at the time of last follow-up if all microbiologic evaluations were negative. Moreover, to make an exhaustive study of proven tuberculosis cases in HIV-seropositive patients, all patients diagnosed with active TB were reviewed to confirm if the case definition was fulfilled.

3.5 STATISTICAL ANALYSIS

Using EPI INFO ver 6.01 and STATISTICA, the following analytic techniques and procedures were carried out:

1. Chi-square test for categorical data and Student's *t* test for continous variables were used for comparing the distributions of sociodemographic factors, risk behaviours, AIDS-defining illnesses, and other variables known to influence the outcome, i.e. potential confounders and effect modifiers. Differences observed were reported with 95 percent confidence interval.

2. Active tuberculosis cumulative incidence was calculated as the number of new cases of active tuberculosis developed during the study period divided by the total number of HIV patients, excluding those diagnosed with active tuberculosis at inclusion.

3. To ascertain the variables predictive of active tuberculosis in HIV-infected patients, logistic regression analysis was performed. In this regression analysis, variables were assessed for significance using stepwise, forward and

backward stepping procedures. Initially, all predictors were forced into the model. Then, in forward selection procedure, predictors were added one by one to the model in the order of the magnitude of their contribution (F to enter statistic corresponding to $p < 0.15$). Variables were not included if they did not exceed the specified F to enter-statistic. Variables stayed in the model once they were included. The backward procedure involved elimination of individual predictors from the full model starting with the variable with the lowest contribution to the model. Elimination stopped when the remaining model exceeded a specified model statistic (F to enter statistics corresponding to $p < 0.1$). In the stepwise procedure, variables were included in the model as in forward selection but at each inclusion the contributions of all the variables in the model at that stage was assessed and elimination occurred as in the backward procedure.

4. The overall survival times for tuberculosis and non-tuberculosis subjects were estimated using the Kaplan-Meier method .The generalized log rank test was used to compare the survival curves.

5. To determine the unadjusted risk of death associated with tuberculosis and to adjust for potential confounding variables, the Cox proportional hazards regression technique was used. Proportionality of the Cox model was checked using the plot of log [-log (survival distribution)] versus log (months) to examine whether survival profiles of the groups included in the model were parallel lines. The models included a priori variables known or thought likely to be associated with outcomes of interest. Age, CD4+ count, history with an

AIDS-defining illness at baseline, and use of preventive and antiretroviral therapy were included as possible confounders and effect modifiers. Ninety-five percent confidence intervals around the risk ratio were calculated using Rothman's appoximation method [2].

6. Similar analysis was done to assess the risk of TB in developing AIDS-defining illness. The Kaplan-Meier method was used for comparing progression to AIDS between TB cases and non-cases who were free of AIDS at inclusion. Log rank method was used to compare the survival curves. The Cox proportional hazards regression method was used to adjust for confounding.

7. To assess whether active tuberculosis disease by itself might have induced a loss of CD4+ counts at the time of active tuberculosis diagnosis, the 3 consecutive CD4+ counts 3-6 months *before, at* and *after* the date of diagnosis of active disease where compared using Friedman and Wilcoxon signed rank nonparametric tests [3]. The choice of using these tests was based on the finding that the CD4+ counts, when tested for normality using Shapiro-Wilks test [3], were significantly non-normally distributed.

8. To avoid using a baseline CD4+ count that might have been influenced by active tuberculosis disease, as the findings of the above analysis may suggest, and which may affect the estimates of the other variables included in the regression model, a further Cox proportional hazards regression analysis was done, including the CD4+ counts 3-6 months *before* or *after,* rather than *at* the

date of diagnosis of active tuberculosis. Using this method, it was possible to estimate parameters for TB and CD4+ lymphocyte count (and other variables included in the model) "adjusted for" for the transient reduction in the CD4+ lymphocyte count caused by active tuberculous disease.

9. To compare the incidence of new AIDS-defining illnesses in TB cases against that of and non-cases, the incidence density ratio (IDR) and 95% confidence intervals were calculated. The incidence density ratio was calculated as the number of AIDS-defining illnesses occurring in each group divided by the total person-months of each group [4]. This analysis was done to obtain an unadjusted incidence density ratio. To adjust for factors known to alter disease progression, the analysis was stratified according to known use of antiretroviral therapy, age and CD4+ count. Adjusted incidence density ratios were calculated using the Mantel-Haenszel formula. Furthermore, the modifying influence of antiretroviral therapy on the effect of active tuberculosis on the incidence of AIDS-defining illnesses was assessed in stratified analysis, using the test for homogeneity of the incidence density ratio across strata [5].

10. The modifying influence of history of AIDS-defining illness and low CD4+ lymphocyte count at inclusion on the effect of active tuberculosis on mortality was assessed in stratified analyses using the Breslow-Day test for homogeneity of the relative risk across strata [6]. Furthermore, the likelihood ratio test was used to test for confounding. The Cox proportional hazards regression was used to assess the significance of interaction between TB and

CD4+ lymphocyte count, and TB and history with AIDS-defining illnesses respectively while adjusting for other predictors of survival. The difference between the log-likelihood of the model containing the interaction term and that of the model without the interaction term was assessed for significance using the chi-square test.

11. To assess the effect of socioeconomic status on survival, a classification method was developed to categorize patients into two mutually exclusive groups of a lower and higher socioeconomic status. This scheme is discussed in appendix III. The Kaplan-Meier method was used for comparing survival of lower and higher demographic status groups. The Log rank method was used to compare the survival curves, and the Cox proportional hazards regression method was used to adjust for confounding.

12. To assess the validity of the WHO [appendix I] and CDC [appendix II] staging systems, survival of cases with tuberculosis was plotted against survival of controls without active tuberculosis. Survival of both groups was measured as time from inclusion to death / censoring or end of the study. Using the WHO and CDC staging systems, controls were staged into one of the 4 stages (WHO) and the 3 stages (CDC) at their inclusion into the study. They were followed from this initial stage till their death or censoring. If a case with tuberculosis presented with an AIDS-defining illness at inclusion, this case was staged in stage 4 (WHO) and stage C (CDC). The Kaplan-Meier method was used to plot the graphs of survival of active tuberculosis group versus other groups with different stages (WHO staging in the first graph and CDC

staging in the second) of HIV infection at inclusion. The Log-rank test was used to test for significance of differences in survival between the TB group and the other groups in the two staging systems. When the survival curve of the TB group overlapped with the survival curve of another group, the prognosis of these two groups was considered similar. Similarity in prognosis was thus the criteria used to judge in which stage of the different stages of the WHO and the CDC HIV staging systems should TB be included.

REFERENCES

1. Centers for Disease Control and prevention, CDC. **Classification system for human T-lyphotropicvirus typeIII/lympadenopathy-associated virus infection 1986**. *MMWR* 35:334-339.
2. Rothman KJ. **Modern Epidemiology** . *New York, Little, Brown and company.* 1986.
3. Armitage P, Berry G. **Statistical methods in medical research**. *London; Blackwell scientific publications*. 1994.
4. Henekens CH, Mayrent S. **Epidemiology in medicine**. *New York; Little Brown and company*. 1987.
5. Sahai H, Khurshid A. **Statistical epidemiology: methods, techniques, and applications**. *New York; CRC press Inc.*1996.
6. Hosmer DW, Lemeshow S. **Applied logistic regression**. *London; Wiley-Interscience Pub. John Wiley and sons.* 1989.
7. Afifi A, Virginia C. **Computer Aided multivariate analysis**. *London; Chapman & Hall*. 1990.

4. RESULTS

Of the 609 HIV patients who enrolled and followed between 1st January 1992 and 31st December 1996, 158 (25.9 %) patients with active tuberculosis met the study inclusion criteria and were enrolled as cases. The comparison group included the remaining 453 (74.1%) HIV-infected patients without active tuberculosis ("controls"). In the general cohort from which the members of this study were drawn, 38 patients were treated for tuberculosis but did not fulfill the case definition and, therefore, were excluded.

4.1 TUBERCULOSIS DIAGNOSIS:

Of the 158 active tuberculosis cases, 90 (57%) presented with pulmonary tuberculosis without evidence of extrapulmonary involvement; 37 (23.4 %) presented with pulmonary tuberculosis and documented evidence of extrapulmonary involvement; 31 (19.6%) presented with extrapulmonary disease only.

Of the 158 TB cases, active disease was confirmed in 69 (43.7 %), 80 (50.6 %) and 9 (5.7%) patients by culture, smear and histologic diagnosis respectively (Fig 4.1).

Fig 4.1

Methods of tuberculosis diagnosis (n=158)

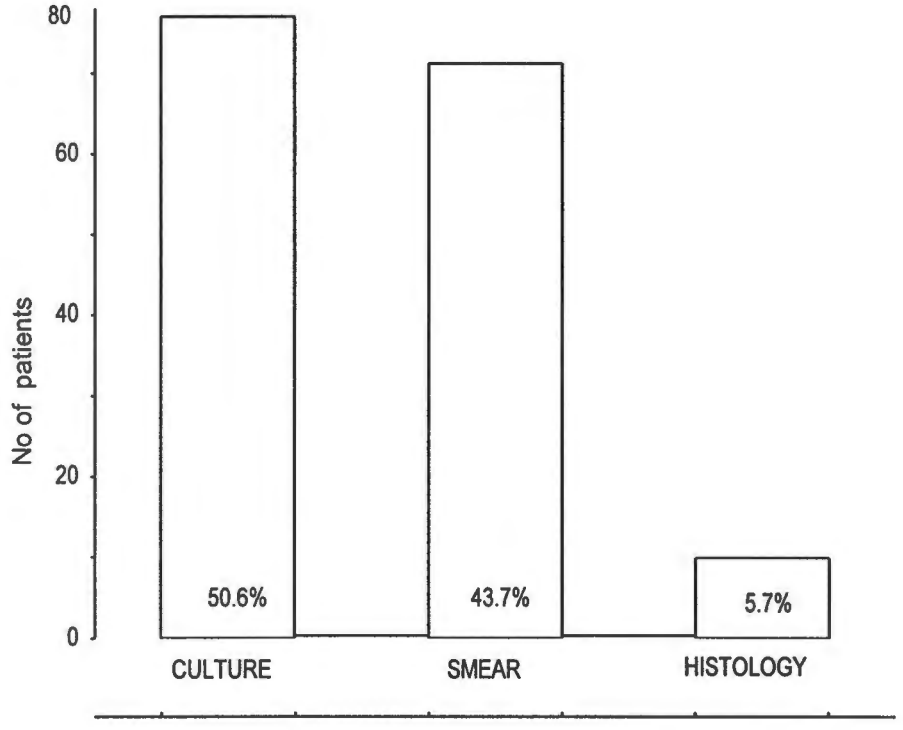


Table 4.1 specifies the source of specimen and method of TB diagnosis. It is clear that sputum was the main source of specimen [115 (72%) cases] in confirming the diagnosis of TB in this cohort.

Table 4.1
Source of specimen used for confirming TB diagnosis (N= 158)

Source	Smear	Culture	Histology	Total	%
Sputum	69	46	-	115	72
Lymph node	-	11	6	17	10.7
Blood	-	4	-	4	2.5
Pleura	-	10	3	13	8.2
Bone marrow	-	1	-	1	0.94
Stool	-	2	-	1	0.94
Urine	-	1	-	1	0.94
Cerebrospinal fluid	-	3	-	3	1.9
Pericardial fluid	-	1	-	1	0.94
Pus	-	1	-	1	0.94
Total	69	80	9	158	100

Patients who were smear (+) and culture (+) were recorded as being culture (+) only.

4.2 GENERAL CHARACTERISTICS AT BASELINE

4.2.1 Gender and age

TB cases and non-cases were similar in gender (Table 4.2.1-a) and age (Table 4.2.1-b).

Table 4.2.1 (a)
Gender of cases and non-cases (N= 609)

Characteristic	Cases (n=158)		Non-cases (n=451)			
	(n)	(%)	(n)	(%)	p-value*	% Difference (C.I.)
Male	79	50	251	58	.09	-8 (-17.0,1.0)

*Chi-square test

Table 4.2.1 (b)
Age distribution of cases and non-cases (N = 609)

Characteristic	Cases (n=158)	Non-cases (n=451)	p-value*
Age (years) , Mean (standard deviation)	32 (9.1)	32 (8.8)	0.9

*Chi-square test

4.2.2 Sexual preferences and risk behaviours

In spite of significantly different proportions of sexual preferences of cases and non-cases across the different categories of sexual preference, heterosexuality predominated in the two groups (Table 4.2.2). None of the comparison group subjects were intravenous drug users, and only one of cases was. The proportions of subjects in the two groups who acquired the disease through contaminated blood transfusion were small relative to other risk factors.

Table 4.2.2
Sexual preferences and risk behaviours of cases and non-cases (N = 609)

Characteristic	Cases (n=158)		Non-cases (n=451)			
	(n)	(%)	(n)	(%)	p-value*	% Difference (C.I.)
Homosexual	10	6.3	88	19.6	.00	-13.3 (-18.6,-8.0)
Heterosexual	125	79	305	67.6	.01	11.4 (3.8,19.0)
IVDU	1	0.6	-	-		-
Blood T/F	2	1.3	7	1.6	.9	-0.3 (-2.4,1.8)
Unknown	20	12.7	51	0.11	.00	12.6 (7.3,17.8)

*Chi-square test T/F: transfusion IVDU: Intra-venous drug users

4.2.3 Distribution by race group

The majority of the patients in the two groups were Black, followed by Coloured, whites and Asian patients. More white patients were reported as non-cases than cases. The proportions of Coloured and Asian patients were not significantly different in the two groups. Black patients were more likely to belong to active tuberculosis group.

Table 4.2.3
Race groups of cases and non-cases (N = 609)

Characteristic	Cases (n=158)		Non-cases (n=451)			
	(n)	(%)	(n)	(%)	p-value*	% Difference (C.I.)
White	10	6.3	95	21	.00	-14 (-20.0,-9.0)
Coloured	30	19	116	25.8	.08	-6.8 (-14.1,0.5)
Black	107	67.7	221	48.8	.00	18.9 (10.3,27.5)
Asian	11	7	19	4.4	.2	2.6 (-1.8,7.0)

*Chi-square test

4.2.4 Marital status

Among the two groups of the study, marital status was not significantly different. Most of the cases and the non-cases were unmarried.

Table 4.2.4
Marital status of cases and non-cases (N = 609)

Characteristic	Cases (n = 158)		Non-cases (n = 451)			
	(n)	(%)	(n)	(%)	p-value*	% Difference
Single	107	67.7	333	73.7	.15	-6 (-14.4,2.4)
Married	33	20.9	93	20.5	.92	0.4 (-6.9,7.8)
Unknown	18	11.4	25	5.8	.02	5.6 (0.21,10.9)

*Chi-square test

4.2.5 Employment status

Unemployment, at time of inclusion into the study, differed significantly within and between the two study groups. Fewer cases were employed compared with non-cases. Of the other known occupation categories.

Table 4.2.5
Employment status of cases and non-cases (N = 609)

Characteristic	Cases (n=158)		Non-cases (n=451)			
	(n)	(%)	(n)	(%)	p-value*	% Difference
Employed	29	18.4	170	37.5	.00	-19.1 (-26.5,-11.6)
Unemployed	97	61.4	181	40	.00	21.4 (12.6,30.2)
Student	9	5.7	29	6	.9	-0.3 (-4.5,3.9)
Pensioner	1	0.6	4	0.9	.8	-0.3 (-18.9,18.3)
Homemaker	2	1.2	11	2	.8	-0.8 (-2.9,1.3)
Disability	2	1.2	8	1.8	.9	-0.6 (-2.7,1.5)
Unknown	18	11.5	50	11.8	.9	-0.3 (-6.1,5.5)

4.2.6 Occupational status

Unskilled patients predominated in both groups. The proportion of unskilled patients was significantly higher in thew case group than the non-case group.

Table 4.2.6
Occupational status of cases and non-cases (N = 609)

Category	Cases (n=158)		Non-cases (n=451)			
	(n)	(%)	(n)	(%)	p-value*	% Difference
Professional/Managerial	4	2.5	30	5.5	.00	-3 (-5.3,-0.7)
White-collar/ Middle-management	3	1.9	35	7.7	.00	-5.8 (-9.1,-2.6)
Skilled/Artisan	5	3.2	27	5	.3	-1.8 (-5.2,1.6)
Semi-Skilled	9	5.7	40	8.8	.2	-3.1 (-7.6,1.4)
Unskilled	137	86.7	319	73	.00	0,20.4)

4.2.7 Number of dependants

In both groups, patients were most likely to have no children /dependants. Very few supported a spouse only. Cases were significantly more likely than non-cases to have one child, and approximately similar proportions of 2 children, or 3 children and more.

Table 4.2.7
Number of dependants of cases and non-cases (N = 609)

Characteristic	Cases (n =158)		Non-cases (n = 451)			
	(n)	(%)	(n)	(%)	p-value*	% Difference (95%C.I.)
Spouse Only	1	0.6	5	1	.4	-0.4 (-1.9,1.1)
1 child	39	24.7	78	17	.03	7.7 (0.1,15.3)
2 children	23	14.6	77	17	.5	-2.4 (-8.9,4.1)
3 children and more	31	19.6	62	13.7	.07	5.9 (-1.1,12.9)
None	64	40.5	229	51.7	.00	-11.2 (-20,1 –2.3)

*Chi-square test

4.2.8 Education level

The patients in both groups were more likely to be uneducated than to have had primary, secondary or tertiary educational level. Non-cases were more likely to have a primary education than TB-cases. However, no significant differences were observed between the two groups at higher levels of education, i.e. secondary and tertiary education.

Table 4.2.8
Education level of cases and non-cases (N= 609)

Characteristic	Cases (n = 158)		Non-cases (n = 451)			
	(n)	(%)	(n)	(%)	p-value*	% Difference (95% C.I.)
Primary	33	20.9	40	8.8	.00	-12.1 (-19.0,-5.0)
Secondary	26	16.5	72	15.9	.87	0.6 (-6.1,7.3)
Tertiary	3	1.9	19	4	.22	-2.1 (-4.9,0.7)
None	75	47.5	190	42	.3	5.5 (-3.5,14.5)
Unknown	21	13.2	130	29.3	.00	-16.1 (-22.9,-9.9)

*Chi-square test

4.2.9 Medical insurance coverage

The majority of patients did not have medical aid scheme coverage. The proportion of non-cases who were members of some medical aid scheme was greater than that of cases.

Table 4.2.9
Medical insurance coverage of cases and non-cases (N = 609)

Characteristic	Cases (n = 158)		Non-cases (n=451)			
	(n)	(%)	(n)	(%)	p-value*	% Difference (95% C.I)
Medical insurance	4	2.5	44	9.7	.00	-7.2 (-10.9,-3.5)

* Chi-square test

4.3 THE CLINICAL CHARACTERISTICS AT BASELINE

4.3.1 Use of antiretroviral therapy and co-trimoxazole prophylaxis

Use of antiretroviral therapy and co-trimoxazole prophylaxis after inclusion into the cohort, differed significantly between the two groups. Zidovudine was the major antiretroviral used in this cohort and was more likely to be used in non-cases. Only 3 non-cases used didanosine. Co-trimoxazole prophylaxis, in contrast, was more likely to be used in cases with TB.

Table 4.3.1
Antiretroviral therapy and co-trimoxazole prophylaxis usage among cases and non-cases (N = 6 09),1992-96

Characteristic	Cases (n = 158)		Non-cases (n = 451)			
	(n)	(%)	(n)	(%)	p-value*	% Difference (95% C.I)
Antiretroviral therapy	3	1.9	44	9.7	.00	-7.8 (-11.3,-43)
Co-trimoxazole	71	44.9	116	25.6	.00	19.3 (10.6,28.0)

*Chi-square test

4.3.2 CD4+ T-lymphocyte count at baseline

At the time of inclusion in the study, cases had significantly lower median CD4+ lymphocyte counts compared to non-cases. A significant difference also existed in the

various levels of CD4+ count with which the patients presented, with cases having a higher proportion in the lowest stratum (≤200 cells /μL) and a lower proportion in the highest stratum (> 400 cells /μL).

Table 4.3.2
CD4+ Count (Cells/μL) at baseline of cases and non-cases (N = 609)

Characteristic	Cases (n=158)		Non-cases (n=451)		p-value	Difference (95% C.I)
CD4 Count						
Median	130		295		0.00	-165 (-245,-86)
Lower quartile	43		158		0.00	-115 (-227.34,-210.66)
Upper quartile	241		460		0.00	-219 (-329,-130)
Minimum	1		1		-	-
Maximum	999		1244		.00	-245 (-329,-130)
	(n)	(%)	(n)	(%)	p-value	% Difference (95%C.I)
≤ 50	43	27	35	7.8	0.00	19.2 (13.4, 25.0)
≤ 200	71	45	149	33	0.00	12 (6.24,17.76)
201-400	45	29	155	34	0.26	-5 (-10.29,0.3)
> 400	42	26	147	33	0.02	-9 (-14.29, -3.71)

4.3.3 AIDS-defining illness at baseline

At inclusion into study, cases presented with AIDS-defining illnesses more frequently than non-cases.

Table 4.3.3
History of AIDS-defining illnesses at baseline in cases and non-cases (N = 609), 1992-96

Disease	Cases (n=158)		Non-cases (n=451)			
	(n)	(%)	(n)	(%)	p-value*	% Difference (95% C.I)
AIDS-defining illnesses	17	10.8	24	5.3	.01	5.5 (1.3,9.8)

*Chi-square test

4.3.4 INCIDENCE OF TUBERCULOSIS

From January 1992 to December 1996, active tuberculosis was reported in 158 patients among the 611 subjects of this study. Of these 158 patients, 113 were diagnosed with active of tuberculosis at the time of their inclusion into the study, and 45 patients were free of active tuberculosis at inclusion but developed active tuberculosis during the course of follow-up. Excluding the 113 patients who were diagnosed with tuberculosis at inclusion, and dividing the remaining 45 patients who developed tuberculosis during the follow-up period by the cohort of 498 who were free of active tuberculosis at inclusion, a cumulative incidence of active tuberculosis of 45 per 498 or 9 percent (or 9036/100,000) during the 5 years can be calculated.

4.3.5 PREDICTORS OF INCIDENT ACTIVE TUBERCULOSIS

To study the factors which were significantly predictive of the occurrence of active tuberculosis in this cohort, and to measure the risk of disease associated with each of these factors, univariate and multivariate logistic regression analyses were carried out. Table 4.3.5 shows the variables which were significantly associated with developing active tuberculosis in this cohort. In univariate analysis, co-trimoxazole prophylaxis was negatively associated with active tuberculosis (p = 0.0043), but not after adjusting for other variables in the multivariate model.

Although not statistically significant in the logistic regression model, the odds of active tuberculosis increased by 1 % percent for every 1-year age increase. It is more meaningful, however, to measure the odds of active tuberculosis disease by taking a 10 year age increase the unit of increment rather than a one year age increase. This

resulted in an odd ratio of 4.67, i.e. a more than a 4-fold increase in the odds of active tuberculosis for every 10year increase in age. Although neither age nor co-trimoxazole prophylaxis were significant in the multivariate model, they were retained owing to their importance in obtaining valid estimates for the other variables in the model.

As shown in the table below, CD4 ≤ 200 cells/μL and a history of AIDS-defining illnesses at inclusion were highly associated with developing active tuberculosis. Although higher education was not significantly associated with active tuberculosis in the univariate model, it emerged as a significantly protective variable against developing active disease after controlling for the other predictors of active disease in the multivariate model. Stage 3 or 4, race (Black or Coloured), and area of residence (lower sociodemographic area) variables were predictive of active disease in both analyses.

It is noteworthy that although stage of disease, level of CD4 count, history of AIDS variables are closely related measures of immunodepression, their simultaneous inclusion in one model is not likely to result in multi-collinearity, as the Cox proportional hazards regression technique is a non-parametric analysis. Thus the estimates provided for these variables in the different multivariate analyses done in this study are valid estimates and are not affected by multi-collinearity.

Table 4.3.5
Predictors of incident tuberculosis (n = 45) in HIV-Infected cohort, 1992-96 (N = 496).

	Univariate models			Multivariate model		
Variable	**OR**	**95%C.I.**	**P**	**OR**	**95%CI**	**P**
Stage 3 or 4	1.96	(1.46,263)	**0.00**	2.07	(1.37,3.11)	**0.00**
CD4≤200 cells/μ L	2.23	(1.2,4.14)	**0.01**	6.79	(1.49,3.89)	**0.01**
History of AIDS	1.22	(0.28,5.39)	**.79**	5.41	(1.04,8,15)	**0.04**
Age>32 years	1.17	(0.63,2.19)	**.61**	1.01	(0.5,2.04)	**0.97**
Co-trimoxazole prophylaxis	0.40	(0.22,0.75)	**0.00**	0.91	(0.42,1.94)	**0.79**
Race group (Black or Coloured)	1.56	(1.02,2.40)	**0.04**	2.24	(1.39,3.60)	**0.00**
Higher education	0.76	(0.56,1.02)	**0.06**	0.71	(0.49,0.99)	**0.04**
Residence (lower socioeconomic area)	2.29	(1.19,4.39)	**0.01**	3.41	(1.58,7.41)	**0.00**

Logistic regression analysis

4.4 MORTALITY

Table 4.4
Mortality in HIV positive cohort, 1992-96

	Cases (n = 158)		**Non-cases (n = 451)**			
	(n)	**(%)**	**(n)**	**(%)**	**p-value**	**% Difference (95% C.I)**
Died	50	32	53	11.7	.00	20.3 (12.2,27.9)

Of the 609 patients 103 (17%) died over the five year period: 50 (31.7%) cases and 53 (11.7%) non-cases. The mortality rate was thus higher in cases than in non-cases (2.8/100 *vs* 0.8/100 person-months; risk ratio (RR) = 2.29 (95% confidence interval (CI) 1.61-3.25).

4.4.1 AIDS-defining illness at baseline:

When stratifying by history of AIDS-defining illnesses, among patients without an AIDS-defining illness a at inclusion, mortality was higher in cases than in non-cases (RR = 3.03, 95 % confidence interval CI 2.04-4.49; $p < 0.001$). In patients with a history of AIDS-defining illnesses at inclusion, mortality was not significantly higher, in cases than in non-cases (RR 1.09, 95 % confidence interval CI 0.63-1.87; $p = 0.77$). This heterogeneity of the RR across the strata of history of AIDS-defining illness at inclusion suggests that the association of active tuberculosis with mortality may be confounded or modified by history of AIDS-defining illness at inclusion. The following Breslow-Day test for the homogeneity of the RR across the strata of AIDS-defining illness at baseline was done to investigate this suggestion.

Table 4.4.1.1
Effect of history with an AIDS-defining illness on the association of TB with mortality.

AIDS-defining illness = Y

TB	Died Y	Died N	
Y	10	7	17
N	13	11	24
	23	18	

RR = 1.09 (0.63,1.87)

P = 0.77

AIDS-defining illness = N

TB	Died Y	Died N	
Y	40	101	141
N	40	387	427
	80	488	

RR = 3.03 (2.04,4.49)

P = 0.000

RR (crude) = 2.69 (1.91,3.79)

RR (Mantel Haenszel) = 2.34 (1.71,3.22)

P = .000

Breslow-Day test for homogeneity of RR across strata

Under the null hypothesis that the RR is constant (homogeneous) across the strata, Breslow-Day test statistic = 4.98, which is significant at one degree of freedom (p = 0.025). Therefore, the hypothesis of the homogeneity of the RR across strata (history with baseline AIDS-defining illness: yes *vs* no) is rejected. Thus a baseline history of AIDS-defining illness modified the effect of active tuberculosis on mortality in this cohort.

The crude and adjusted (Mantel-Haenszel) estimates of the RR were also slightly different, indicating some confounding. This was further investigated using a Cox proportional hazards regression model [Table 4.4.1.2].

Table 4.4.1.2
Modelling the effect of history of an AIDS defining illness on the association between TB and mortality

Model	Log-likelihood	Log-likelihood Difference*	P*
1- Tuberculosis	-370.218		
2- Tuberculosis + history with AIDS	-368.157	2.061	.15
3- Tuberculosis + history with AIDS + Interaction term (Tuberculosis * history with AIDS)	-361.698	6.459	0.01

Cox proportional regression hazards model
* Chi-square test of significance

From the above table, the difference in log-likelihood statistics between the first model (the model containing TB alone) and the second model (the model containing TB and history of AIDS) (2.06) is not significant on Chi-square test with one degree of freedom (p = 0.15). This indicates that despite the difference between the crude and

adjusted RR on the previous analysis, history of an AIDS-defining illness at baseline is a not a confounder of the effect of TB on mortality.

After introducing the interaction term, the difference in the log-likelihood statistic (6.49) between the second model (the model containing TB and history of an AIDS-defining illness) and the third model (the model containing TB, history with an AIDS-defining illness and the interaction term), was significant (p = 0.01). This confirms that history of an AIDS-defining illness at baseline is an effect modifier of the effect of active tuberculosis on mortality, in agreement with the Breslow-Day test for homogeneity of the RR in stratified analysis.

4.4.2 CD4 + T-lymphocyte count at baseline:

When stratifying by level of CD4+ count at baseline, among patients with CD4+ lymphocyte count > 200 cells /µl at inclusion, mortality was significantly higher in cases than in non-cases (RR = 2.94, 95% CI 1.94 – 4.54; p = 0.005). However, this significant difference was not observed among patients with CD4+ lymphocyte count ≤ 200 cellsµ/l at baseline. In this stratum, mortality was higher, but not significantly, in cases than in non-cases (RR = 1.39, 95 % CI 0.74 – 2.62; p = 0.2). The following test were done to test for confounding/effect modification of CD4+ count ≤ 200 cells /µL of the association between active TB with mortality

Table 4.4.2.1
Effect of CD4+ count ≤ 200 cells /μL on the association of active TB with mortality.

CD4+ count ≤ 200 cells / μL

TB	Died Y	Died N	
Y	14	57	71
N	20	129	149
	34	186	

RR = 1.39 (0.74,2.62)

P = 0.40

CD4+ count > 200 cells / μL

TB	Died Y	Died N	
Y	36	51	87
N	33	269	302
	69	320	

RR = 2.94 (1.94,4.54)

P = 0.000

RR (crude) = 2.29 (1.61,3.25)

RR (Mantel Haenszel) = 2.29 (1.62,3.23) *P* = .001

Breslow-Day test for homogeneity of RR across strata :

Under the null hypothesis that the RR is constant (homogeneous) across the strata, the Breslow-Day test statistic = 11.28, which is highly significant at one degree of freedom (chi-square value at one degree of freedom = 3.84). Therefore, the hypothesis of the homogeneity of RR across the two CD4+lymphocye strata is rejected. The high estimate of the test statistic reflects the degree of heterogeneity of the RR across the strata of CD4+ lymphocyte count. Thus, CD4+ lymphocyte at baseline modifies the effect of active tuberculosis on mortality in this cohort. Furthermore, the crude and adjusted (Mantel-Haenszel) estimates of the RR are similar, indicating that CD4+ lymphocyte ≤ 200 at baseline is not a confounder. This was further investigated using a Cox proportional hazards regression model [Table 4.4.2.2].

Table 4.4.2.2
The effect of CD4+ count ≤ 200 cells /μL on the association of active TB with mortality

Model	Log-likelihood	Log-likeli-hood Difference	P*
1- Tuberculosis	-370.218		
2- Tuberculosis + CD4+ count ≤ 200 cells /μL	-368.548	1.67	.196
3- Tuberculoss + CD4+ count ≤ 200 cells /μL + Interaction term (Tuberculosis * CD4+ count ≤ 200 cells /μL)	-361.245	7.30	.006

Bivariate Cox proportional hazards regression models
*P-value for Wald Chi-square

In the above table, the difference in the log-likelihood statistics between the first model (the model containing TB alone) and the second model (the model containing TB and CD4+ count ≤ 200 cells /μL) (1.67) is not significant (p = 0.196). This indicates that CD4+ count ≤ 200 cells /μL at baseline is a not a confounder of the effect of TB on mortality. After introducing the interaction term, the difference in the log-likelihoods between the second and the third model (the model containing TB, CD4+ count ≤ 200 cells /μL and the interaction term) (7.303) was significant (p = 0.006). confirms that CD4+ count ≤ 200 cells /μL at baseline is an effect modifier of the effect of active tuberculosis on mortality; in agreement with the Breslow-Day test for homogeneity of the RR in stratified analysis.

The combined effects of these two variables and other predictors of mortality on the association between active tuberculosis and mortality in multivariate analysis is presented later.

4.5 FOLLOW-UP AND SURVIVAL

Follow-up in the two groups was similar (mean = 11.5 and 14.6 months in cases and non-cases respectively; $p = 0.7$). The proportion of patients lost to follow-up in the two groups was not significantly different [59 (37%) cases *vs* 153 (34%) non-cases; $p = 0.5$].

The Kaplan-Meier survival distributions indicate that active tuberculosis cases had an overall shorter survival than non-TB controls. The overall median survival was 23.9 months for prevalent cases (cases with active TB at inclusion), and 25 months for incident cases (cases free of TB at inclusion but who developed active disease later), compared with 47.7 months for non-cases (Fig 4.5.1). Survival of non-cases was significantly different from that of prevalent cases ($p < 0.001$), and from incident cases ($p < 0.001$) of active tuberculosis. No significant difference in survival time between prevalent and incident cases of active tuberculosis was observed, ($p = 0.8$).

Fig 4.5.1
Survival of cases and non-cases (N = 609)

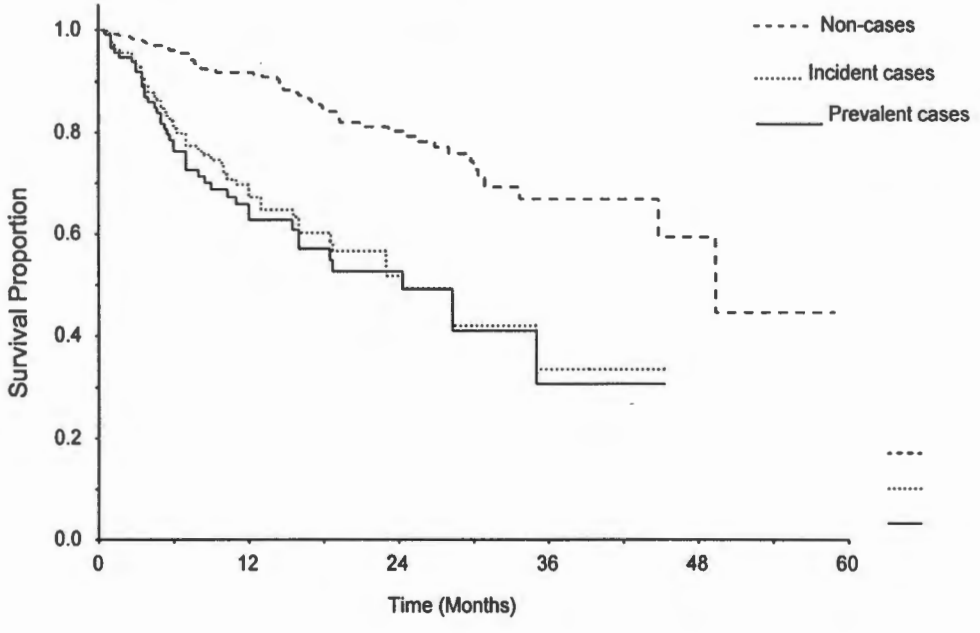
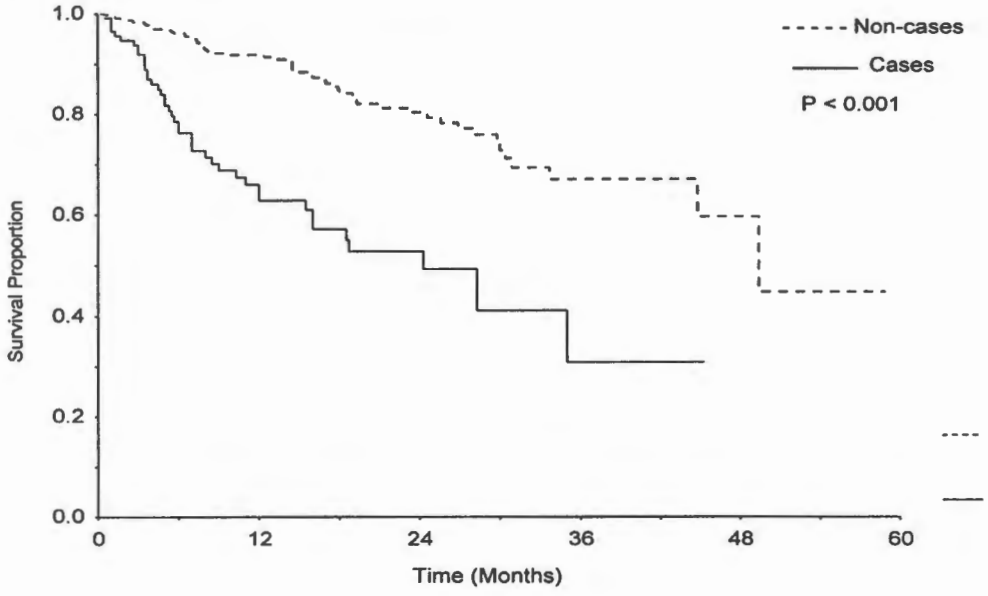


Fig 4.5.2
Survival of cases and non-cases (N = 609)



The difference in survival between the prevalent and incident cases of active tuberculosis was small. When all TB cases combined were compared to non-cases survival times were significantly different ($p < 0.001$) (Fig 4.5.2).

Table 4.5.1
Cumulative probability of survival according to study group and length of survival (N = 609)

	Cumulative probability of survival		
	1 Year	2 Year	3 Year
	(percent ± S.E.)		
Cases	69 ± .04	49 ± .08	28 ± 0.2
Non-cases	92 ± .02	82 ± .03	64 ± .08

S.E : standard error

The Kaplan-Meier probabilities of survival (Table 4.5.1) shows that the 1-year survival proportion for all cases was consistently less than the 1-year survival for non-

cases (69% versus 92%); the same relationship was present at 2 years (49% versus 82%, cases *vs* non-cases) and 3 years (28% and 64%) respectively.

Stratified analyses were done to investigate survival differences between cases and non-cases at various levels of CD4 + lymphocyte counts. A significant difference in survival between cases and non-cases (42 cases *vs* 147 non-cases) existed at CD4+ lymphocyte count > 400 cells /μ L (Fig 4.5.3) and CD4+lymphocyte count 201- 400 cells/μ L (45 cases *vs* 155 non-cases) (Fig 4.5.4).

Fig 4.5.3
Survival of cases and non-cases with CD4+ > 400 cells μ/l at inclusion (n =219)

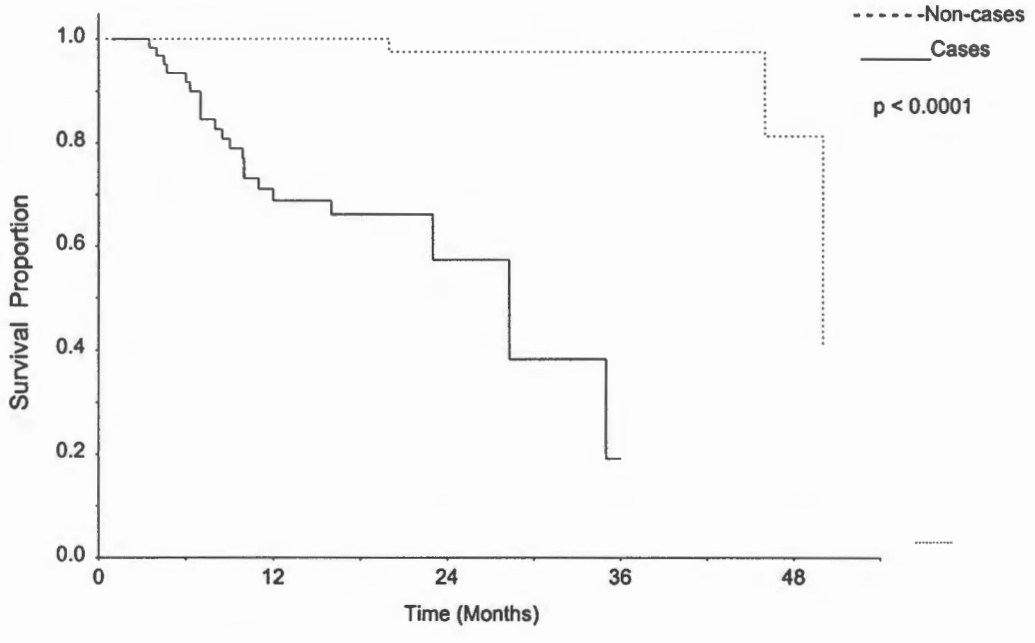
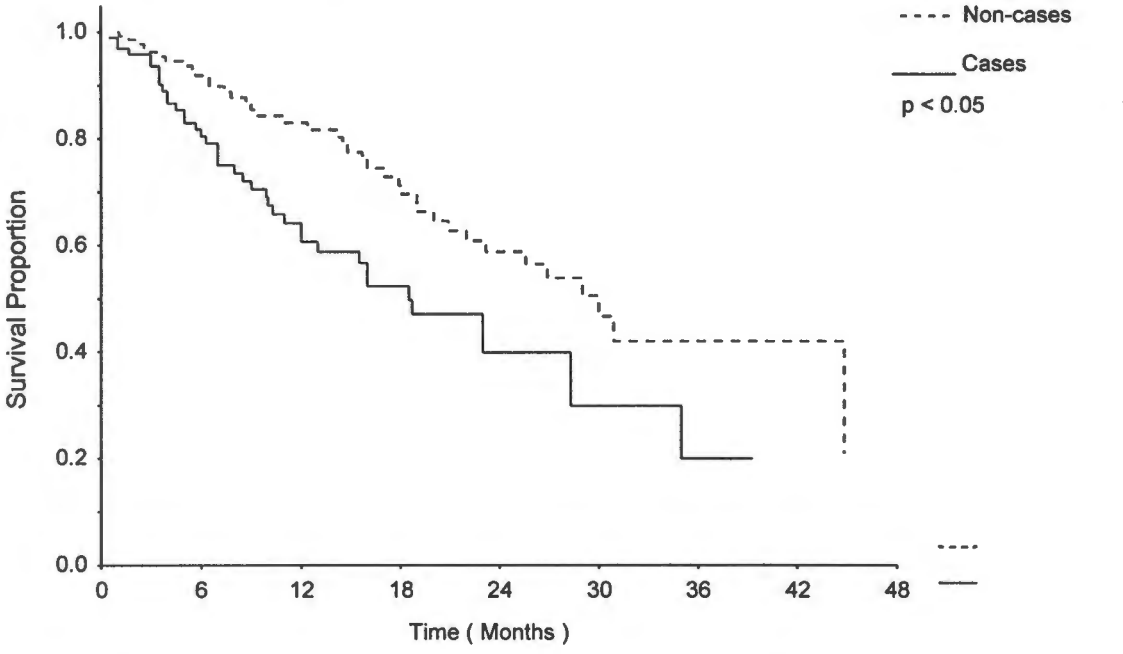


Fig 4.5.4
Survival of cases and non-cases with CD4 count 201-400 cells/μ L at inclusion (n = 200)



However, when stratifying survival by patients with CD4 + count ≤ 50 cells/μl (43 cases *vs* 35 non-cases) (Fig 4.5.5), or CD4 + count ≤ 200 cells/μl at inclusion (71 cases *vs* 149 non-cases) (Fig 4.5.6), or history of an AIDS-defining illness (17 cases *vs* 24 non-cases) (Fig 4.5.7), no significant difference in survival was observed between the two groups.

Fig 4.5.5
Survival of cases and non-cases with CD4 + count ≤ 50 cells μ/l at inclusion (n = 78)

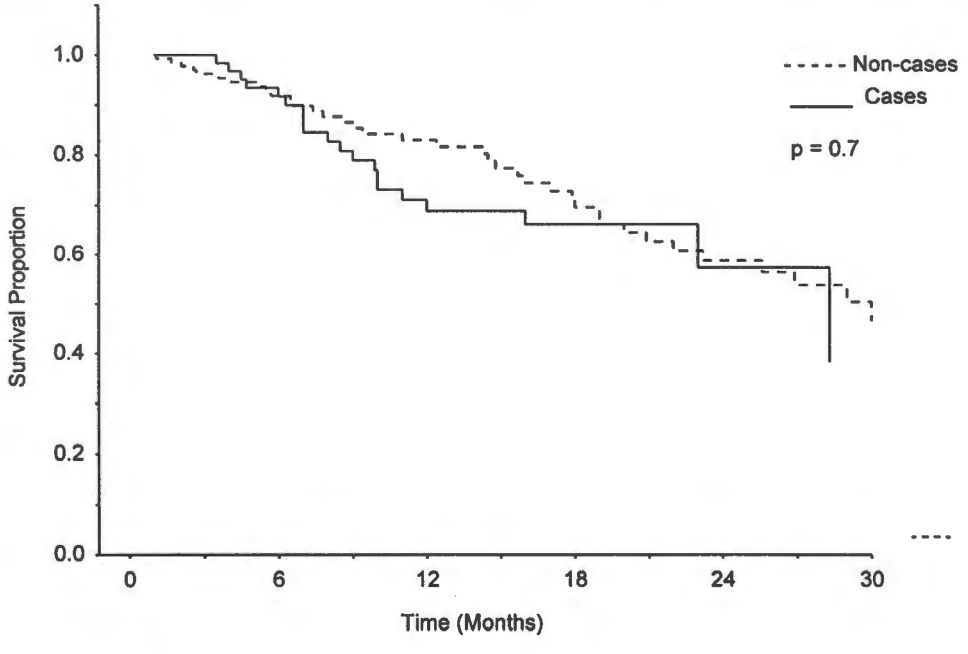


Fig. 4.5.6
Survival of cases and non-cases with CD4 + count ≤ 200 cells μ/l at inclusion (n = 220)

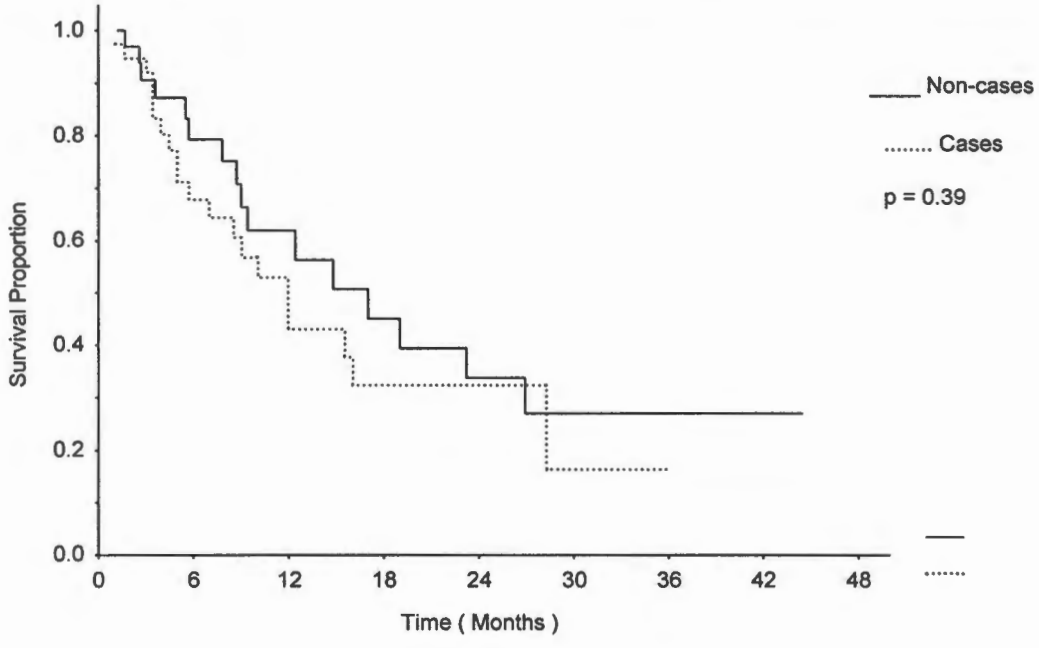
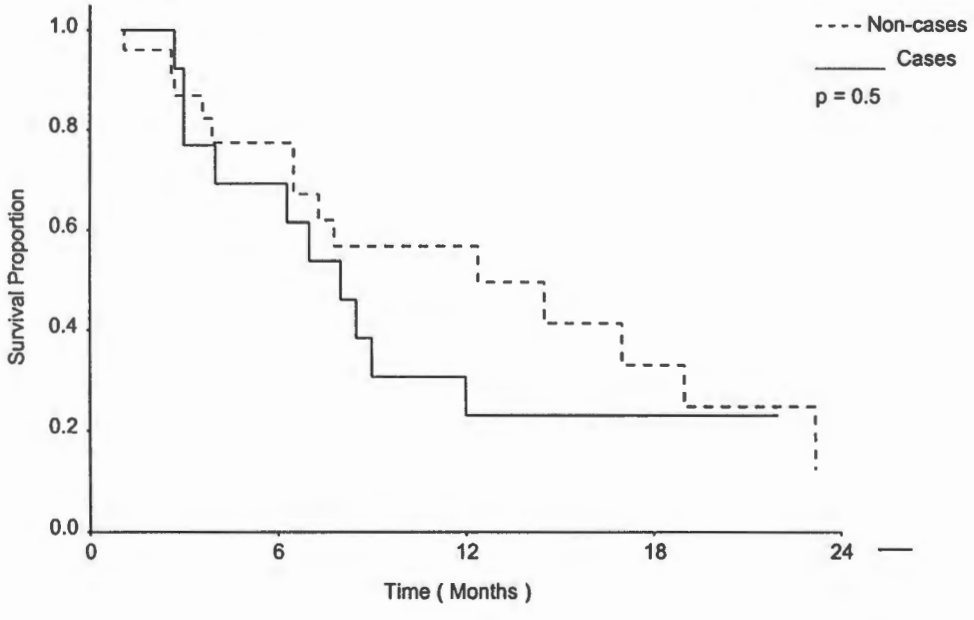


Fig 4.5.7
Survival of cases and non-cases with an AIDS defining illness at inclusion (n = 41)

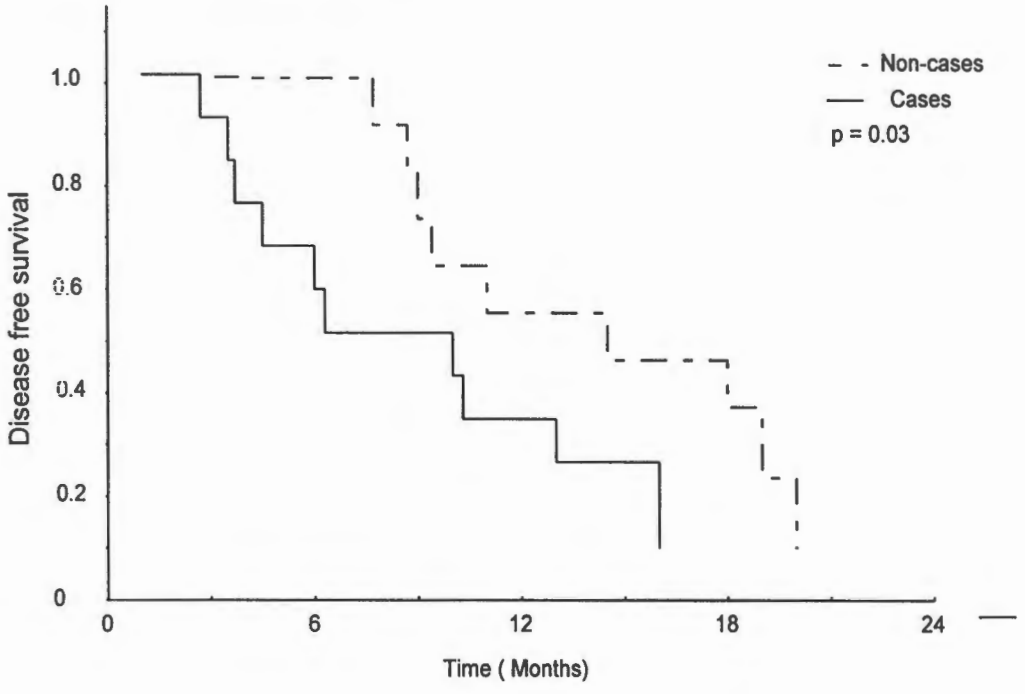


4.6 Progression to AIDS

Time to progression to AIDS was compared between cases and non-cases. In cases and non-cases who were free of AIDS at time of inclusion into study but developed AIDS during follow-up (33 cases *vs* 34 non-cases), a significant difference was observed (Fig 4.6). HIV-infected patients with active tuberculosis were diagnosed with an AIDS-defining illness; on average after 6 months, compared with an average of 14.5 months for HIV-infected patients without active tuberculosis.

Fig 4.6

Time to AIDS of cases and non-cases who were AIDS-free at inclusion (n = 76)



To adjust for the factors known to influence the effect of TB to the progression to AIDS, the following multivariate Cox proportional hazards regression was done. It is clear that even after controlling for age, CD4 count, antiretroviral therapy and co-trimoxazole prophylaxis, TB was an independent risk

Table 4.6
Predictors of AIDS in TB cases and non-cases; multivariate analysis

Variable	Beta	S.E	RR	RR 95% CI	p
Age >32	0.0168	0.1767	1.01	(0.71,1.44)	**0.9**
CD4 < 200 cells/μL	0.8613	0.2019	2.36	(1.59,3.50)	**0.00**
Tuberculosis	0.4763	0.2056	1.61	(1.08,2.41)	**0.02**
Co-trimoxazole prophylaxis	-.5363	0.2179	0.21	(0.14,0.33)	**0.00**
Antiretrovirals	-0.0086	0.0109	0.99	(0.97,1.01)	**0.4**

Cox proportional hazards regression analysis. S.E: Standard error. CI : confidence interval

4.7 INCIDENCE OF AIDS-DEFINING ILLNESS

In the general cohort, 76 (13.38%) of the 568 patients without an AIDS-defining illness at baseline developed at least one AIDS-defining illness. These patients developed 90 AIDS-defining illnesses over 8422.1 person-months of observation, yielding an incidence density of 1.1 infections per 100 person-months. In the 158 TB cases, a total of 47 AIDS-defining illnesses occurred in 33 patients over 1803.4 person-months of observation, yielding an incidence density of 2.6 AIDS-defining illnesses per 100 person-months. In contrast, in the 451 non-cases, a total of 43 AIDS-defining illnesses occurred in 34 patients over 6618.7 person-months, yielding an incidence density of 0.7 infections per 100 person-months.

The crude incidence density ratio of cases to non-cases of active tuberculosis for AIDS-defining illnesses was thus 3.7 (95% confidence interval CI 2.85-6.28); risk ratio (RR) = 4.10 (95% confidence interval 2.65-6.07).

To assess whether there were factors other than active tuberculosis operating in the greater frequency of AIDS-defining illnesses observed in active tuberculosis cases relative to non-tuberculosis controls, a stratified analysis was done. Table 4.7.1 shows that incidence densities of AIDS-defining illnesses were consistently greater in cases than non cases across the different stratum of age, use of antiretroviral therapy and CD4+ count.

Table 4.7.1
Incidence of AIDS-defining illnesses in TB cases and non-cases (1992-96), stratified by age, use of antiretroviral therapy and CD4 + count ≤200

Variable	Group	(n)*	Person months	Incidence** density	IDR (95%CI) p-value	Adjusted IDR
Over all	Cases	47	451.7	10.4	1.5 (1.02,2.32)***	
	Non-cases	43	635	6.8	$p = 0.04$	
Age ≤ 32	Cases	21	260.8	8.1	1.6 (0.74,3.19)	
	Non-cases	11	210	5.2	$p = 0.24$	1.71 (1.12,2.0)
Age >32	Cases	26	190.9	13.6	1.8 (1.08,3.03)	
	Non-cases	32	425	7.5	$P = 0.022$	
Antiretroviral used	Cases	2	47.7	4.2	0.4 (0.1,1.9)	
	Non-cases	14	144	9.7	p = 0.25	1.58 (1.03,2.0)
No antiretroviral used	Cases	45	404	11.0	1.9 (1.18,3.0)	
	Non-cases	29	491	5.9	$p = 0.007$	
CD4+ Count ≤200 cells/μ L	Cases	31	249.8	12.4	1.7 (1.03,2.78)	
	Non-cases	31	421.5	7.4	$P = 0.037$	1.59 (1.06,2.0)
CD4+ Count >200 cells/μ L	Cases	16	201.9	7.9	1.4 (0.67,2.98)	
	Non-cases	12	213.5	5.6	$P = 0.37$	

*n= number of AIDS-defining illnesses; **(n) per 100 person-months; IDR= Incidence density ratio
***Adjusted IDR ; Mantel-Haenszel test

However in the stratum who used antiretroviral therapy, the incidence of AIDS-defining illnesses was lower (but not significantly) in cases than non-cases. This suggested that use of antiretroviral therapy might have modified the effect of tuberculosis on the incidence of AIDS-defining illnesses in this cohort. This was further investigated in the following analysis.

Table 4.7.2
Effect of antiretroviral therapy on the association of TB on the incidence of AIDS-defining illnesses.

	Antiretroviral therapy used		Antiretroviral therapy not used	
	No.*	person-months	No.*	person-months
Cases	2	47.7	45	404
Non-cases	14	144	29	491
	IDR = 0.43		IDR = 1.9	
	95% CI = (0.1-1.9)		95% CI = (1.18-3.0)	
	$P = 0.25$		$P = 0.007$	

Adjusted incidence density ratio = 1.58
95% CI = (1.03-2.0)
$p < 0.05$

Test for homogeneity of incidence density ratio across strata *No.: Number of AIDS defining illnesses

Under the null hypothesis that the incidence density ratio (IDR) is constant (homogeneous) across the strata, the test statistic = 0.6629. From the Chi-square table, $p(\chi_1^2 > 0.6629) = 0.416$. Thus the hypothesis of the homogeneity of IDR for the two strata is rejected. This test suggests that antiretroviral therapy, in the stratum who used it, eliminated the effect of TB in increasing the incidence of AIDS-defining illnesses. The effect of TB in increasing the incidence of AIDS-defining illnesses was ,however, apparent in the stratum who did not use antiretroviral therapy. Thus

antiretroviral therapy modified the effect of active tuberculosis on the incidence of AIDS-defining illnesses in this cohort.

4.8 PREDICTORS OF MORTALITY

To control further for differences at baseline and known predictors of mortality in HIV-infection, a Cox proportional hazards regression analysis was carried out. Below are the variables which were significantly associated with survival. Although the variables age and antiretroviral therapy were not significantly associated with mortality in this cohort, they were retained in the model owing to their importance in modelling survival of patients with tuberculosis and HIV infection.

Table 4.8.1
Predictors of mortality in cases and non-cases, 1992-96: univariate analyses

	Beta	Standard Error	Risk Ratio	RR 95% C.I	p
CD4≤200 μ/l	.4047	.2490	1.50	(0.92,2.44)	**.10**
Antiretroviral therapy	-.233	.2557	0.79	(0.48,1.31)	**.36**
Co-trimoxazole prophylaxis	.3593	.1994	0.70	(0.51,0.95)	**.07**
Tuberculosis	.7173	.2033	2.05	(1.38,3.07)	**.00**
Age > 32 years	.0368	.2003	1.04	(0.70,1.54)	**.85**
AIDS-defining illness	.5570	.2378	1.75	(1.10,2.78)	**.01**

Cox proportional hazards regression models

Table 4.8.2
Predictors of mortality in cases and non-cases, 1992-96 : multivariate analysis

	Beta	Standard Error	Risk Ratio	RR 95% C.I	p
CD4≤200 µ/l	.7436	.2823	2.10	(1.22,3.66)	.00
Antiretroviral therapy	.2582	.3024	1.29	(0.72,2.34)	.39
Co-trimoxazole prophylaxis	-.916	.2452	0.40	(0.25,0.65)	.00
Tuberculosis	.8644	.2489	2.37	(1.46,3.86)	.00
Age> 32 years	.2587	.2133	1.30	(0.85,1.97)	.22
AIDS-defining illness	.8227	.2605	2.28	(1.36,3.78)	.00

Cox proportional hazards regression model

Table 4.8.3

Predictors of mortality in cases and non-cases, 1992-96: univariate and multivariate analysis compared

	Univariate Model			Multivariate Model		
Variable	**RR**	**95% C.I**	**P**	**RR**	**95% C.I**	**P**
CD4≤200 µ/l	1.50	(1.92,2.44)	.10	2.10	(1.22,3.66)	.00
Antiretroviral therapy	0.79	(0.48,1.31)	.36	1.29	(0.72,2.34)	.39
Co-trimoxazole prophylaxis	0.70	(0.51,0.95)	.05	0.40	(0.25,0.65)	.00
Tuberculosis	2.05	(1.38,3.07)	.00	2.37	(1.46,3.86)	.00
Age > 32 years	1.04	(0.70,1.54)	.85	1.30	(0.85,1.97)	.22
AIDS-defining illnesses	1.75	(1.10,2.78)	.01	2.28	(1.36,3.78)	.00

Cox proportional hazards regression analyses.

In the multivariate Cox proportional hazards regression analysis that simultaneously controlled for a number of predictors, it is evident that even after controlling for differences in baseline characteristics and known predictors of mortality in HIV

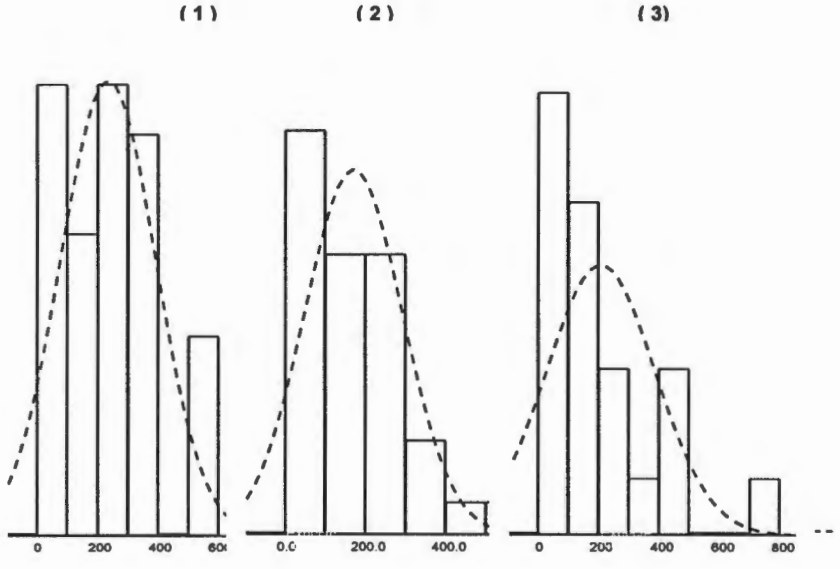
infection, active tuberculosis conferred an independent risk for death in HIV-infected patients.

In this model, CD4 counts ≤ 200 cells/μl, active tuberculosis and presence of an AIDS-defining illness at baseline were significantly associated with poorer survival. Co-trimoxazole prophylaxis was consistently protective against death. Specifically, the risk of death increased approximately two fold for a CD4 count ≤ 200 cells / μL, a diagnosis of active tuberculosis and a previous history of AIDS-defining illness. Co-trimoxazole prophylaxis decreased risk of death by 60% in those who used it compared to those who did not. Use of antiretroviral therapy and greater age were not significant predictors of mortality in this cohort.

4.9 EFFECT OF TUBERCULOSIS ON CD4+ T-LYMPHOCYTE COUNT

This section sets out to assess to what extent active tuberculosis by itself induces a loss in CD4+ count by determining (1) the effect of using a CD4 + count *before* or *after* rather *at* TB diagnosis, and (2) the impact of TB on CD4 count.

Fig 4.9.1
Test of normality for CD4+ T-lymphocyte count before, at and after the date of tuberculosis diagnosis (incident cases only, n = 45)

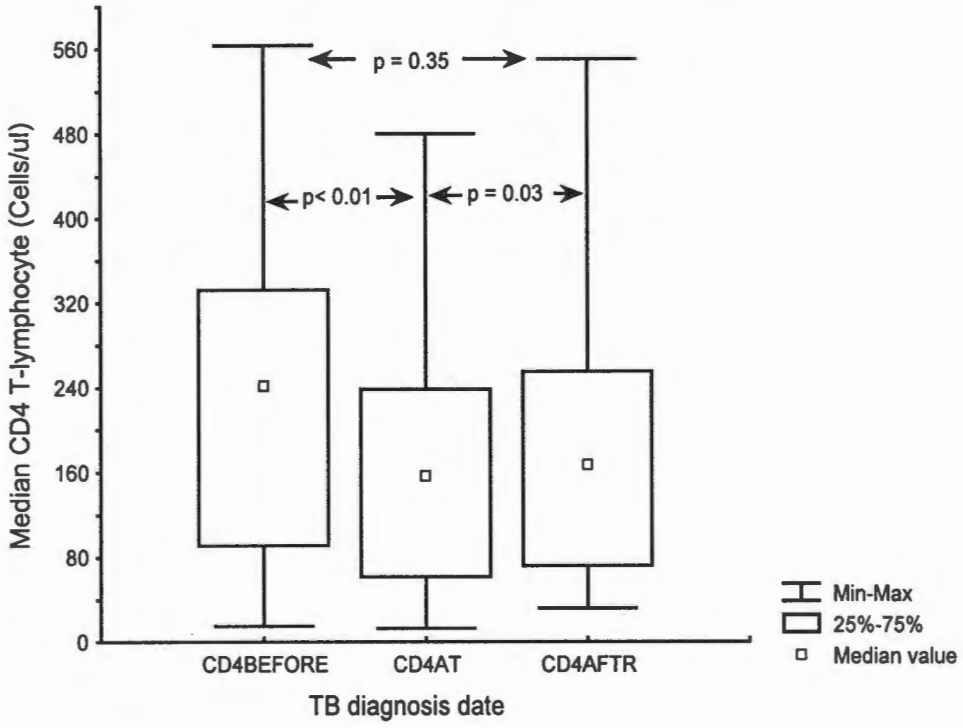


Shapiro-Wilk's test of normality

(1) Before TB diagnosis date (p<.03) (2) At TB diagnosis date (p<.04) (3) After TB diagnosis date (p <.001)

CD4+ counts of the 45 incident active tuberculosis cases ± 3-6 months *before, at* and *after* the date of a confirmed diagnosis of active disease were compared (Fig 4.9.2, Table 4.9.1). Non-parameteric analysis based on Friedman nonparametric Anova analysis (Table 4.9.1) was done. The choice of this test was based on the finding that CD4+ counts were not normally distributed (Fig 4.9.1).

Fig 4.9.2
Median CD4 + lymphocyte count *before, at* and *after* TB diagnosis (n = 45)



Further comparisons between the 3 CD4+ counts were done using Wilcoxon Match Pairs Test (Table 4.9.2). From this table it is evident that the CD4+ counts 3-6 months *before, at* and *after* tuberculosis diagnosis were significantly different (p = 0.03). However, the CD4+ counts 3-6 months *before* compared to those 3-6 months *after* active tuberculosis diagnosis date were not significantly different (p = 0.35).

The significantly different values (p = 0.002) of the CD4+ counts *before* and *at* active tuberculosis diagnosis date reflect the substantial loss in CD4+ count which active tuberculosis disease induced in HIV-infected patients of this cohort. Partial recovery in CD4+ count after active tuberculosis diagnosis was apparent in the significant difference (p < 0.03) in CD4+ count *at* compared to that *after* active tuberculosis diagnosis date.

Table 4.9.1
Difference between CD4 counts *before, at*, and *after* TB diagnosis date (incident cases , n = 45)

Variable	Average rank	Mean	S.D	Median	Q_{Lower}	Q_{Upper}
CD4 BEFORE TB	2.25	234.81	156.41	242	91	333
CD4 AT TB	1.59	176.7	136.27	157	62	239
CD4 AFTER TB	2.15	205.72	180.02	167	72	255

Friedman ANOVA and Kindall Coefficient of concordance (the ANOVA Chi Sqr. (N = 45, df = 2) = 6.675676 **p < 0.03553** Coefficient concordance = .15172 Aver. Rank r = 0.11133

Friedman nonparametric Anova analysis. S.D: standard deviation
Q_{Lower} : Lower quarile Q_{Upper} :Upper quartile

Table 4.9.2
Comparisons between CD4 counts *before, at*, and *after* TB diagnosis date (incident cases, n=45)

PAIR OF VARIABLES	VALID N	p
CD4 AT TB *vs* CD4 BEFORE TB	45	**0.00**
CD4 AT TB *vs* CD4 AFTER TB	45	**0.03**
CD4 BEFORE TB *vs* CD4 AFTER TB	45	**0.35**

Wilcoxon nonparametric Matched Pairs Test

To account for the apparent loss in CD4+ count which active tuberculosis might have induced in the 45 incident and the 113 prevalent cases of active tuberculosis of this cohort, another multivariate analysis was carried out. In the following Cox proportional hazards regression model, the CD4+ count 3-6 months *before* or *after* active tuberculosis diagnosis date (for the 45 *incident* cases) and 3-6 months *after* active tuberculosis diagnosis date (for the 113 *prevalent* cases) were included in the model, rather than the CD4+ counts *at* diagnosis date, as in the previous analyses.

Table 4. 9.3
CD4 count (3-6) months before or after TB diagnosis date and association with mortality (N = 158): univariate analysis

	Beta	Standard Error	Risk Ratio	R R 95% C.I.	p
CD4 ≤ 200	.9078	.4086	2.48	(1.11,5.53)	**.02**

Cox proportional hazards regression model

Table 4.9.4
CD4 count (3-6) months before or after TB diagnosis date and other predictors of mortality : multivariate analysis

	Beta	Standard Error	Risk Ratio	RR 95%CI	p
CD4≤200	.6086	.2817	1.84	(1.06,3.19)	**.03**
Antiretriviral therapy	.0653	.3202	1.07	(0.57,1.99)	**.83**
Co-trimoxazole	-.7564	.2483	0.47	(0.29, 0.76)	**.00**
Tuberculosis	.5328	.2472	1.70	(1.05,2.77)	**.03**
AIDS-defining illnesses	.8277	.2589	2.29	(1.38,3.79)	**.00**
Age>32 years	.2077	.2129	1.23	(0.81,1.87)	**.32**

Cox proportional hazards regression model

Table 4.9.5
CD4 count (3-6) months before or after TB diagnosis date and other predictors of mortality (N=158): univariate and multivariate analyses compared

	Univariate Model			Multivariate Model		
Variable	**RR**	**95% C.I**	**P**	**RR**	**95% C.I**	**p**
CD4≤200 µ/l	2.48	(1.11,5.53)	**.02**	1.84	(1.06,3.19)	**.03**
Antiretroviral therapy	0.79	(0.48,1.31)	**.36**	1.07	(0.57,1.99)	**.83**
Co-trimoxazole	0.70	(0.51,0.95)	**.05**	0.47	(0.29,0.76)	**.00**
Tuberculosis	2.05	(1.38,3.07)	**.00**	1.70	(1.05,2.77)	**.03**
Age > 32 years	1.04	(0.70,1.54)	**.85**	1.23	(0.81,1.87)	**.32**
AIDS-defining illnesses	1.75	(1.10,2.78)	**.01**	2.29	(1.38,3.78)	**.00**

Cox proportional hazards regression model

After controlling for differences at baseline in known predictors of survival in HIV-infected patients and the depressed level of CD4+ T-lymphocyte count at tuberculosis diagnosis, active tuberculosis increased mortality by 70% in HIV-infected patients compared to HIV-infected patients without active tuberculosis.

Table 4.9.6
Predictors of mortality in cases and non-cases including (1) CD4 count at TB diagnosis and (2) CD4 count 3-6 months *before* or *after* TB diagnosis : multivariate analyses

	(1)			(2)		
Variable	**RR**	**95% C.I**	**P**	**RR**	**95% C.I**	**P**
CD4≤200 µ/l	2.10	(1.22,3.66)	**.00**	1.84	(1.06,3.19)	**.03**
Antiretroviral therapy	1.29	(0.72,2.34)	**.39**	1.07	(0.57,1.99)	**.83**
Co-trimoxazole prophylaxis	0.40	(0.25,0.65)	**.00**	0.47	(0.29,0.76)	**.00**
Tuberculosis	2.37	(1.46,3.86)	**.00**	1.70	(1.05,2.77)	**.03**
Age > 32 years	1.30	(0.85,1.97)	**.22**	1.23	(0.81,1.87)	**.32**
AIDS-defining illnesses	2.28	(1.36,3.78)	**.00**	2.29	(1.38,3.78)	**.00**

Cox proportional hazards regression analyses

The multivariate analyses shown in the table above, indicate that the predictive model remained essentially the same using CD4 count *at* or *before* or *after* date of TB diagnosis. The risk ratio associated with the active tuberculosis and the CD4 count were reduced somewhat after adjustment, but remained significant predictors of mortality.

4.10 Interactions between CD4+ lymphocyte count, baseline AIDS-defining illnesses and tuberculosis in the presence of other predictors of mortality

The Breslow-Day and log-likelihood tests done in a previous section showed that both CD4+ lymphocyte ≤200 cells/μL and history of AIDS-defining illness were effect modifiers of the association of active tuberculosis with mortality. As the parameter estimates of the variables included in the model may change in the presence of interaction between these two variables and tuberculosis, it was important to test the significance of these interactions in the presence of other predictors of mortality included in the Cox proportional hazards regression model. Interaction terms were created by multiplying each of the two variables with tuberculosis and were included in the model. The difference between the log-likelihood of the two multivariate models (*before* and *after* including the interaction terms) was tested for significance.

Table 4.10.1
Testing the significance of the interaction terms.

Model	Interaction term	Log-likelihood	Difference	P*
1	Starting model (table 4.28)	-503.30		
2	AIDS-defining illness * TB	-500.943	2.36	.125
3	CD4+lymphocyte ≤200 cells/μL*TB	-500.867	2.43	.121
4	CD4+lymphocyte ≤200 cells/μL* AIDS-defining illness *TB	-499.185	4.116	.128

Multivariarte Cox proportional hazards regression models *P Chi-square test

Model 1 (the starting model [Table 4-28]) included the variables antiretroviral therapy, co-trimoxazole prophylaxis, CD4+ lymphocyte count ≤ 200 cells/μL, tuberculosis, AIDS-defining illness and age > 32 years at baseline. Model 2 included all these variables plus the interaction term between AIDS-defining illness and

tuberculosis. Model 3 included the variables in the starting model plus the interaction term between baseline CD4 lymphocyte count ≤ 200 Cells/μ L and tuberculosis. Model 4 included the variables in the starting model plus the interaction term between baseline CD4 lymphocyte count ≤ 200 Cells/μ L, AIDS-defining illness and tuberculosis.

Form the above table it is clear that none of the interaction terms were significant, i.e. they were rejected by the multivariate Cox proportional hazards regression model. This is despite the fact that both history of AIDS-defining illness and CD4 lymphocyte count (≤ 200 Cells/μ L) at inclusion where shown in sections 4.4.1 and 4.4.2 to be effect modifiers and their interaction with TB was shown to be significant. This suggests that the heterogeneity of the RR across the strata of CD4 lymphocyte count (≤ 200 and > 200 Cells/μ L) shown previously in the Breslow-Day stratified analysis (section4.4.2); can not be attributed to the modifying effect of CD4 lymphocyte count ≤ 200 Cells/μL at baseline to the association of TB with mortality alone, but rather to the modifying effects of the other variables (antiretroviral therapy, co-trimoxazole prophylaxis, AIDS-defining illnesses at inclusion or age > 32) not control for in that stratified analysis. The interaction between the two variables was attenuated after adding the effects of other predictors of survival operating in the increased association between active tuberculosis and mortality. This may explain why the interaction term was rejected by the multivariate model. The same interpretation applies to the rejection of the interaction between the baseline AIDS-defining illnesses and tuberculosis in the multivariate model.

In conclusion, from all the analyses carried out above, active tuberculosis emerged as an independent predictor of death in HIV-infected patients after controlling for known predictors of mortality in HIV infection; and after examining for interaction effects.

4.11 EFFECT OF CO-TRIMOXAZOLE ON MORTALITY AND MORBIDITY OF HIV-INFECTED PATIENTS WITH TB

To specifically investigate the effect of co-trimoxazole prophylaxis on the survival of HIV-infected patients with active tuberculosis, stratified univariate and multivariate analyses were done. In this cohort, co-trimoxazole prophylaxis was started in patients with a CD4+ count lower than 200 cells/μL. Survival of 71 HIV-infected patients with active tuberculosis and reported use of co-trimoxazole prophylaxis was compared with that of the remaining 87 HIV-infected patients with active tuberculosis patients for whom use of co-trimoxazole prophylaxis was not reported during follow-up (controls).

The incidence of AIDS-defining illnesses which could be prevented by co-trimoxazole (*Pneumocystis carinii* pneumonia, cerebral toxoplasmosis, non-typhoid salmonellosis and bacterial pneumonia) was lower in the co-trimoxazole treated group (3 patients; incidence density = 0.3/100 person-months) than in the untreated group (14 patients; incidence density = 2.1/100 person-months; risk ratio = 0.23; 95 % confidence interval = 0.06-0.84; $p = 0.017$).

Mortality was also lower in the co-trimoxazole treated group (14 patients; 1.2/100 person-months) compared to the untreated group (36 patients; 5.5/100 person-months; $p = 0.002$). The median survival (Fig 4-11) of the co-trimoxazole treated group was

longer than the untreated group (28 versus 16 months; p = 0.03). In the univariate analysis (Table 4.11.1), use of co-trimoxazole prophylaxis improved survival (risk ratio = 0.53; 95% confidence interval = 0.30-0.94; p = 0.03). A similar protective effect was found after adjustment for differences at baseline (risk ratio = 0.38; 95% confidence interval = 0.23-0.62; $p < 0.001$) in the multivariate analysis (Table 4.11.2). It is noteworthy that the better outcome in patients given co-trimoxazole prophylaxis was achieved despite their lower CD4+ lymphocyte count at baseline than that of patients not given the treatment (median 174 cells/µl, interquartile range 33-199 compared to 308 cells/µl, interquartile range 290-745; $p<0.001$).

Table 4.11.1
Effect of co-trimoxazole on survival of HIV-infected patients with tuberculosis, 1992-96 (N =158)

	Beta	Standard Error	Risk Ratio	RR 95% C.I..	p
Co-trimoxazole	-.6299	.2908	0.53	(0.30,0.94)	**.03**

Cox proportional hazards regression univariate model (including only patients with active TB)

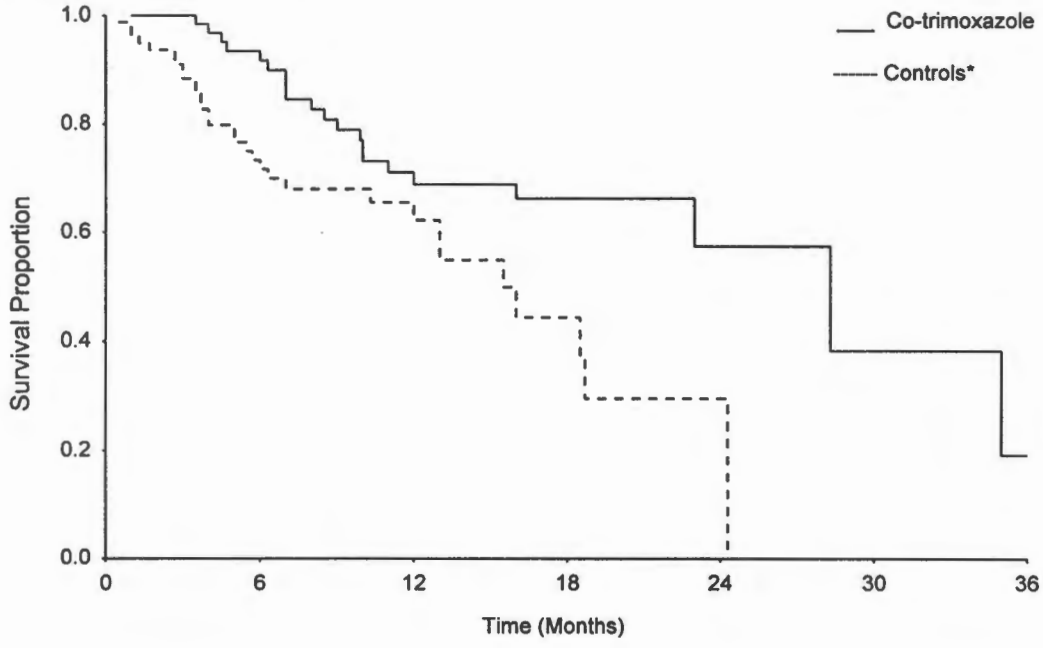
Table 4.11.2
Effect of co-trimoxazole on survival of HIV-infected patients with TB controlling for other predictors of mortality, 1992-96(N =158)

	Beta	Standard Error	Risk Ratio	RR 95%CI	p
CD4≤200 cells/µL	1.0873	.3491	2.96	(1.49,5.87)	**.00**
Co-trimoxazole	-.9711	.2474	0.38	(0.23,0.62)	**.00**
AIDS-defining illnesses	.7902	.2708	2.20	(1.29,3.74)	**.00**
AGE>32	.0912	.2181	1.09	(0.72,1.68)	**.67**

Cox proportional hazards regression multivariate model (including only patients with active TB)

Patients with active tuberculosis who used co-trimoxazole prophylaxis survived significantly longer compared to tuberculosis patients who did not used co-trimoxazole prophylaxis (median = 28 months versus 16 months respectively, p = 0.030) (Fig 4.11).

Fig 4.11
Effect of co-trimoxazole on survival of HIV-infected patients with TB



*Controls: HIV-infected patients with TB who did not receive co-trimoxazole because their CD4+ count was >200 cells/μL

4.12 EFFECT OF SOCIOECONOMIC STATUS ON SURVIVAL

To investigate the effect of socioeconomic status on survival, a further stratified analysis was done. Patients were categorized into two groups; higher socioeconomic and lower socioeconomic patients. The first group was chosen as a reference group. Employment status, occupation status, education level, medical aid coverage, number of dependents and residence in an area characterised by lower socioeconomic status (as defined by the Cape Town Metropolitan Council, CMC- Appendix V) and

medical aid scheme coverage were used as surrogates for socioeconomic status. This scheme eventually resulted in the inclusion of all patients in white race group in this cohort in the higher socioeconomic group, and all other race groups in this cohort in the lower socioeconomic group. Thus, race group and socioeconomic status in the analyses done in this section will be used interchangeably for assessing the effect of race or socioeconomic status on survival in this cohort. Survival of higher socioeconomic status (50 months) was significantly greater than that of patients in the lower socioeconomic status/non-white race group (30 months); ($p < 0.001$) (Fig 4.12.1). This difference in survival was similar to that observed when patients were categorized according to race group (Fig 4.12.2).

Fig 4.12.1
Effect of socioeconomic status on survival of HIV-infected patients, 1992-96 (N = 609)

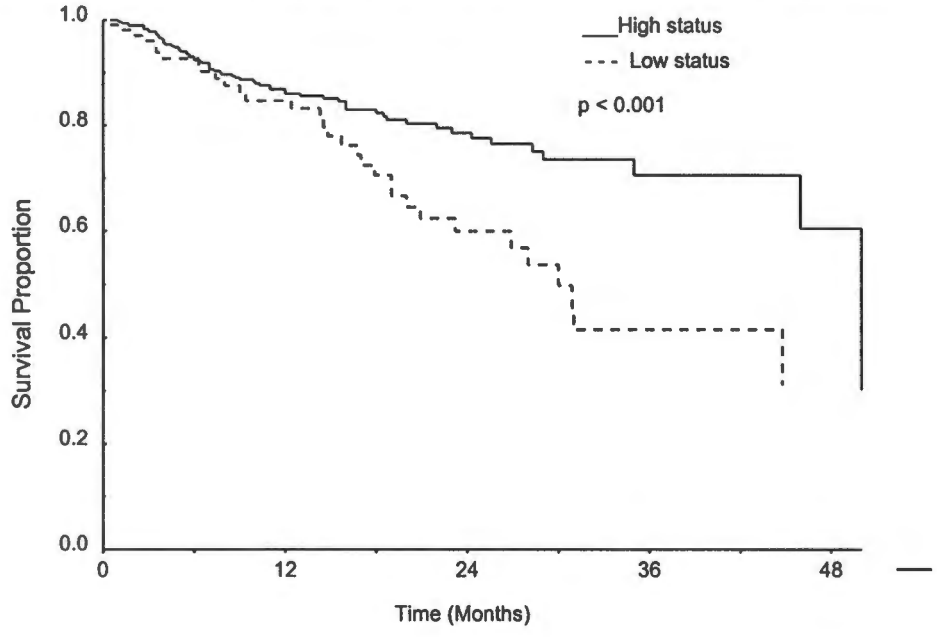
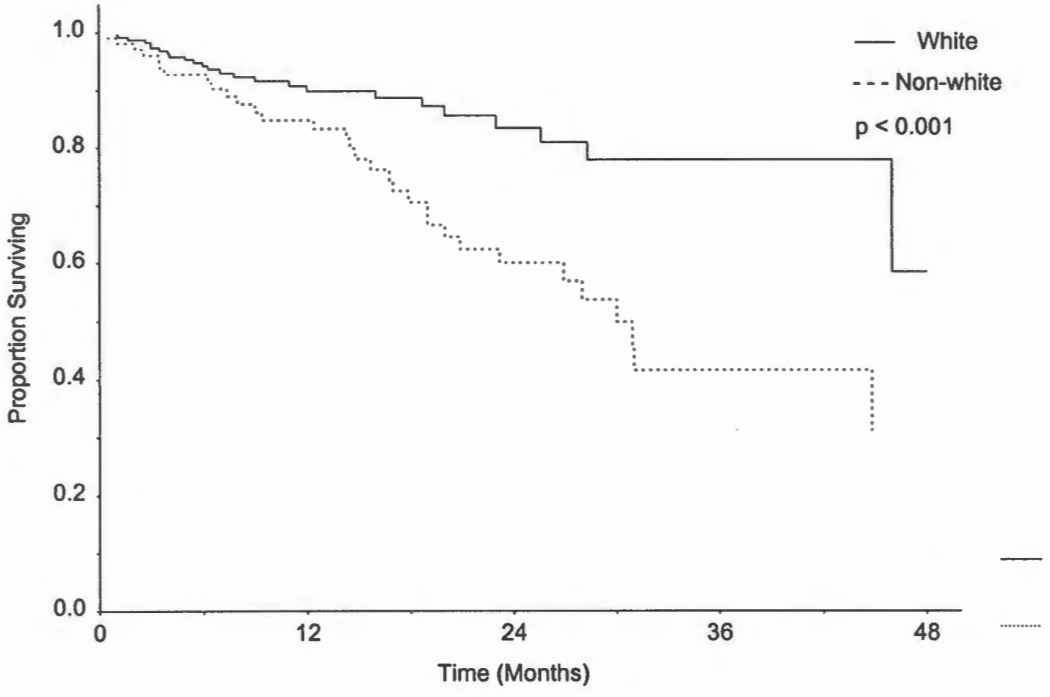


Fig 4.12.2
Effect of race group on survival of HIV-infected patients, 1992-96 (N = 609)



To adjust for other predictors of survival that might have contributed to the observed differences in survival between groups, further univariate and multivariate analysis were carried out controlling for baseline CD4+ counts, use of antiretroviral therapy, co-trimoxazole prophylaxis, age, presence of AIDS-defining illness at baseline, and active tuberculosis. In the univariate model, higher socioeconomic status was protective against mortality (RR = 0.64, 95% confidence interval 0.42-0.99, $p = 0.04$) (Table 4.12.1).

Table 4.12.1
Effect of socioeconomic status on survival of HIV-infected patients (N = 609)

	Beta	Standard Error	Risk Ratio	RR 95% C.I.	p
Higher Socioeconomic Status	-.4412	.2175	0.64	(0.42,0.99)	.04

Univariate Cox proportional hazards regression model

However, after adjusting for other predictors of mortality, no significant difference was observed between the two groups (RR = 0.80, 95% confidence interval 0.42-1.57; p = 0.53). This indicates that the significant difference in survival observed in the above univariate analysis could perhaps be attributed to differences in medical care (*viz* co-trimoxazole prophylaxis), level of presenting immunodeficiency (*viz*, CD4 T-lymphocyte count) and severity of disease or co-morbidity (TB, AIDS-defining illness), rather than race and socioeconomic differences as shown in Table 4.12.2 below.

Table 4.12.2
Effect of socioeconomic status on survival of HIV-infected patients controlling for other predictors of mortality (N = 609)

	Beta	Standard Error	Risk Ratio	RR 95% C.I	p
Antiretroviral therapy	.2771	.3554	1.31	(0.66,2.65)	**.43**
Co-trimoxazole prophylaxis	-.7870	.2625	.45	(0.27,0.76)	**.00**
CD4≤ 200 /μL	.7401	.3432	2.09	(1.07,4.09)	**.03**
Tuberculosis	.7074	.2618	2.02	(1.22,3.39)	**.00**
AIDS-defining illnesses	.7996	.2626	2.22	(1.33,3.73)	**.00**
Age>32 years	.3162	.2248	1.37	(0.88,2.13)	**.15**
Higher socioeconomic status	-.2132	.3399	.80	(0.42,1.57)	**.53**

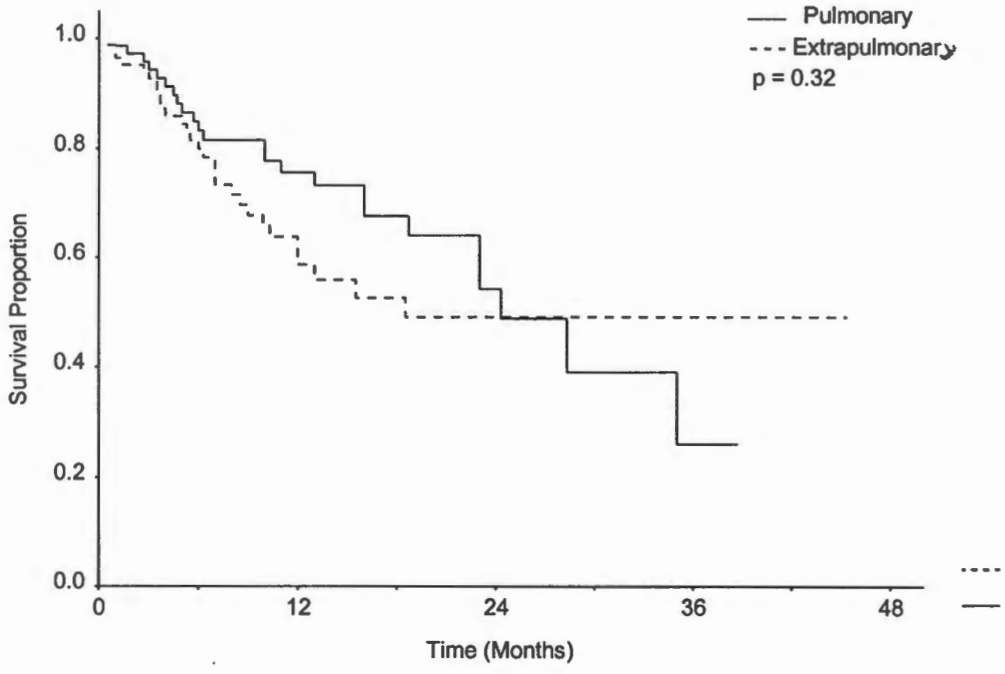
Cox proportional hazards regression model multivariate analysis.

4.13 EFFECT OF SITE OF DISEASE ON SURVIVAL

To study the effect of site of active tuberculosis on survival of HIV-infected patients, a stratified survival analysis was done. Patients diagnosed with active tuberculosis were further stratified according to site of disease into pulmonary and extrapulmonary

groups. Fig 4.13.1 shows no significant differences ($p = 0.32$) in survival between the two groups.

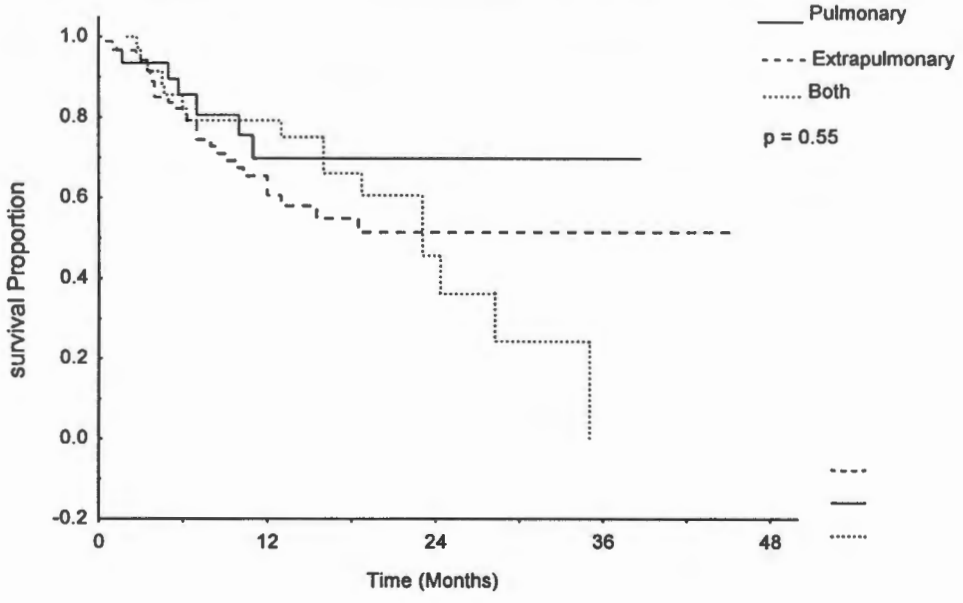
Fig 4.13.1
Survival of active tuberculosis patients according to site of disease, 1992-96 (N =158)



The same non significant difference was observed when the two groups were further stratified into patients diagnosed with pulmonary, extrapulmonary or both (Fig 4.13.2) ($p = 0.55$).

Fig 4.13.2

Survival of active tuberculosis patients according to site of disease



4.14 VALIDITY OF HIV STAGING SYSTEMS IN HIGH TB PREVALENCE SETTING

The prognostic utility of the clinical criteria of the WHO and CDC staging systems was assessed in this cohort drawn from a high tuberculosis prevalence setting. Controls (i.e. non-TB cases) who never developed active tuberculosis during follow-up were staged into the above two different staging schemes at their inclusion into study. Their respective survival times curves from inclusion into the study and the initial stage they presented with until death or censoring were plotted against the survival curve of patients with active tuberculosis.

Using the WHO staging system, controls with stage 1 or stage 2 where combined in one group as their survival was similar (Fig 4.14.1). Survival of patients with active tuberculosis was not significantly different from survival of patients presented with stage 3 ($p = 0.25$), but was significantly better than survival of patients who presented with stage 4 (or AIDS) ($p = 0.000$) (Fig 4.14.2). This suggests that HIV-infected patients with active tuberculosis and no other opportunistic infections have a prognosis comparable to that of other HIV-infected patients with no active tuberculosis but diagnosed with any of the conditions of WHO stage 3.

In the CDC staging system, the survival curve of patients with active tuberculosis coincides with the survival curve of patients who presented with stage B disease (Fig 4.14.3).

The survival of patents with active tuberculosis only (without other conditions of WHO stage 3 or 4, CDC stage B or C) was further compared with the survival of patients without active tuberculosis but with other conditions of WHO stage 3, CDC stage B. Only patients diagnosed with one of the WHO stage 3, CDC stage B conditions were included in this stratified survival analysis. The survival for these individual conditions were examined to determine which of these curves was asymptotic to the survival curve of the active tuberculosis group. Survival of the active tuberculosis group was found to be similar to survival of oral hairy leukoplakia and oral candidiasis groups (WHO stage 3, CDC stage B). Survival of these two groups was combined into one group and compared with survival of the active tuberculosis group. Fig 4.14.4 indicates that in HIV-infected patients with active tuberculosis survival is similar to that of patients diagnosed with oral hairy leukoplakia and/or oral candidiasis with no active tuberculosis (and no any of WHO stage 3 or 4, CDC stage B or C conditions).

Fig 4.14.1
Survival of controls with stage 1 vs stage 2 at inclusion (WHO staging system)

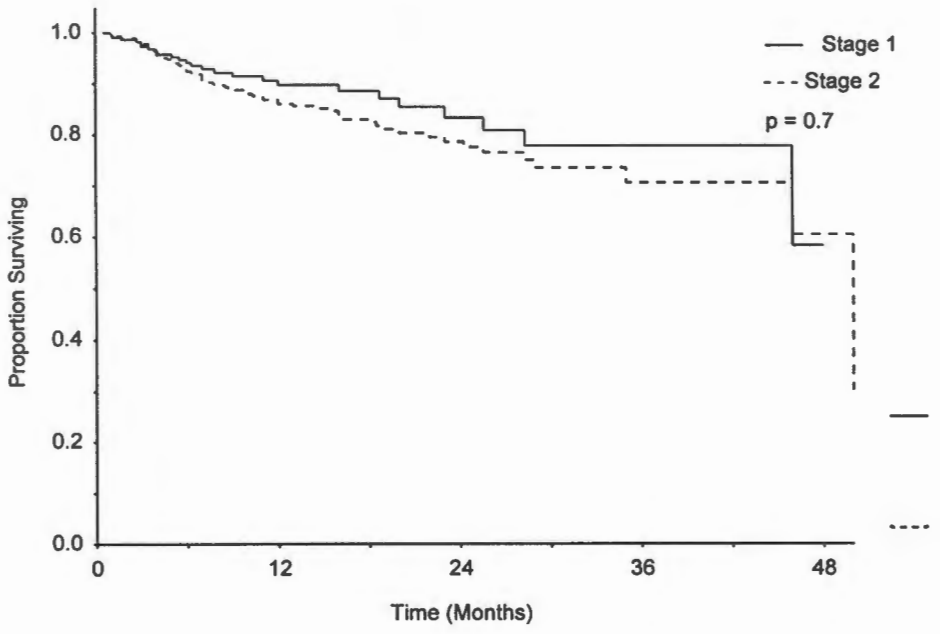


Fig 4.14.2
Patients with active TB *vs* patients with other conditions (WHO staging criteria) (N=609)

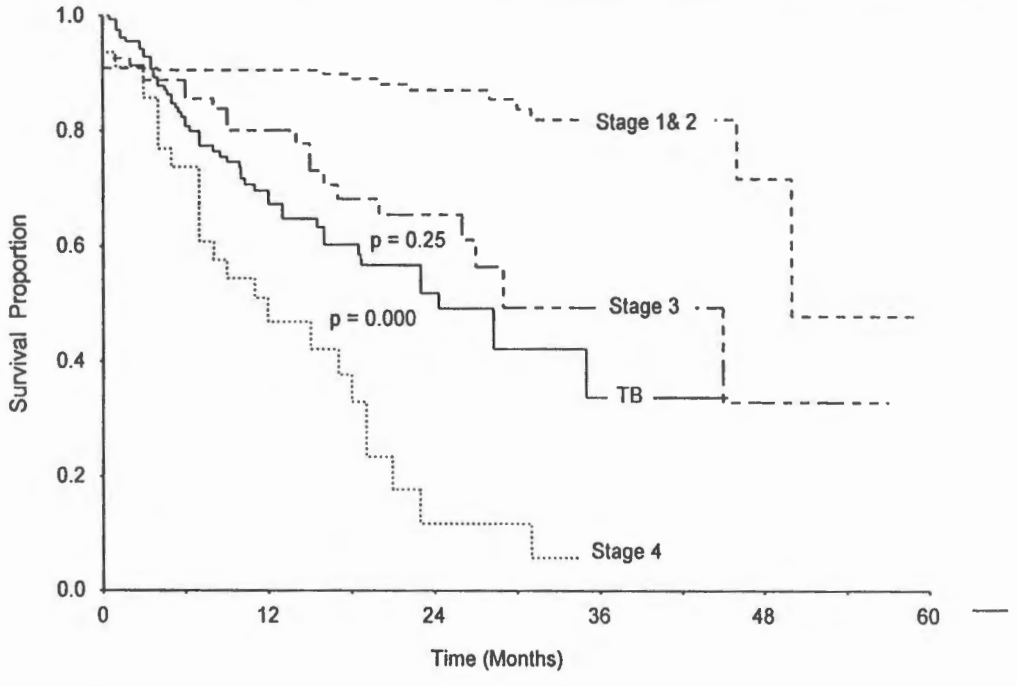


Fig 4.14.3
Patients with active TB *vs* patients with other conditions (CDC staging criteria) (N= 609)

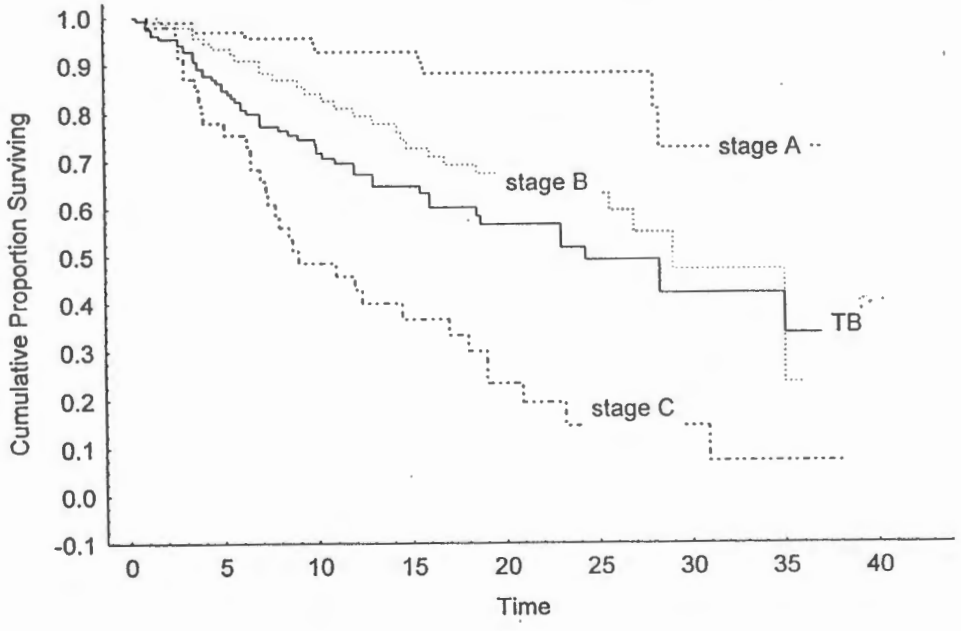
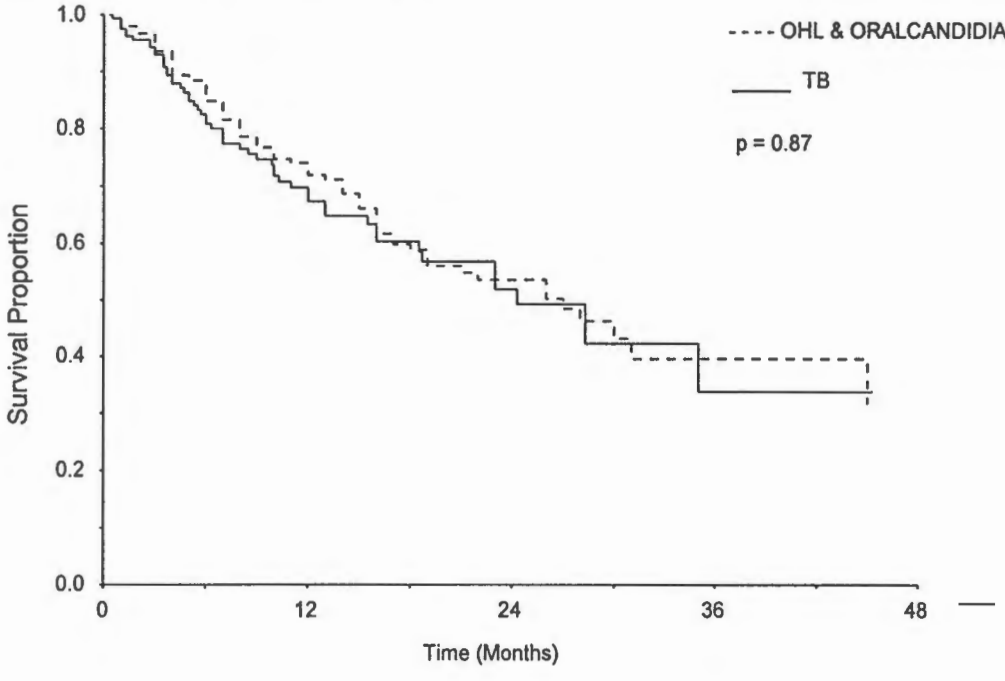


Fig 4.14.4
HIV-infected patients with active TB *vs* HIV-infected patients with oral hairy leukoplakia or candidiasis (WHO stage CDC stage B)



5. DISCUSSION

This prospective cohort study was undertaken to assess the prognostic effect of active tuberculosis on the natural history of human acquired immunodeficiency syndrome in patients presenting to the two major HIV clinics in Cape Town by comparing the survival experience of a group of 158 HIV-infected patients with active tuberculosis with that of a comparison group of 451 HIV-infected patients without active tuberculosis.

The findings of this study reflect the magnitude of the deleterious effect which active tuberculosis has on the prognosis of HIV-infected patients. Being done in a setting with one of the highest recorded tuberculosis prevalences in the world, this study, by documenting the increased incidence of AIDS-defining illness and the reduced survival observed in HIV-infected patients with active tuberculosis, illustrates dramatically the link between HIV infection and active tuberculosis.

5.1 SURVIVAL OF HIV-INFECTED PATIENTS WITH ACTIVE TUBERCULOSIS.

5.1.1 Effect of tuberculosis on mortality

In this cohort, HIV-infected patients diagnosed with active tuberculosis had a reduced survival when compared to HIV-infected patients without active tuberculosis. This difference in survival between the two groups was consistent at higher levels of presenting CD4 lymphocyte count (201-400 cells/μL or > 400 cells/μL). However, this significant difference in survival associated with tuberculosis was not observed when patients presented with an AIDS-defining illness or a very low CD4+ lymphocyte count (≤ 200 cells/μl or ≤ 50 cells/μl) at the time of inclusion into the study. This may be explained by fact that these patients were already terminally ill, with advanced immunosuppression, making it impossible to detect a significant difference in survival based on tuberculosis status.

The reduced survival of HIV-infected patients with active tuberculosis observed in this study is in agreement with studies done elsewhere [1,2]. Leroy *et al.* observed that when survival between the two groups (i.e. HIV-infected patients with and without tuberculosis) was compared according to the presence of AIDS-defining illness at inclusion, there was no significant difference in survival between the two groups [1]. In contrast, a significant difference in survival between the two groups was observed in the Whalen *et al.* study, where survival was reduced in tuberculosis cases compared with that in controls subjects in patients (1) with AIDS-defining illness at baseline [$p = 0.001$]; and (2) in patients without AIDS-defining illness [$p =$

0.001] [2]. However, non of these studies compared the survival of patients according to presenting level of CD4+ lymphocyte count.

The median survival time of HIV-infected patients with active tuberculosis in this cohort is comparable to that reported in developed countries: *viz.* the USA (21-30 months) [3,4], and Europe (22 months)[5]. It is, however, significantly greater than that reported in sub-Saharan African countries: e.g. Rwanda (15 months) [6]. The significantly greater survival of HIV-infected patients with active tuberculosis in this cohort than those from Rwanda may be due to differences in the management of patients, *viz.* the wide use of rifampicin and co-trimoxazole prophylaxis in this cohort compared to other African countries were neither of these interventions is widely used.

Pozniak *et al.* Have argued that rifampicin "is a good bactericidal drug with a potent sterilizing effect, and the ability to prevent the emergence of drug resistance. In addition to acting on rapidly dividing bacilli, it kills so-called "persisters", which remain inactive for long periods but have intermittent periods of metabolism, with only short drug exposure. This is crucial to its sterilizing ability, and also confirms experimental studies of TB of both early bactericidal activity and and sterilizing activity"[7].

Co-trimoxazole prophylaxis is an effective intervention for improving survival and reducing the incidence of the AIDS-defining illnesses frequently diagnosed in HIV-infected patients from African countries, such as salmonellosis, isosporiasis, cerebral toxoplasmosis, and bacterial pneumonia. Evidence of its efficacy in Africa is lacking,

it has been thought to be suitable for HIV-infected patients from developed countries where the spectrum of AIDS-defining illnesses is different. A total of 71 (45%) TB patients in this cohort had co-trimoxazole prophylaxis, which was observed to increase survival and reduce mortality (see section 5.3).

The improved survival in this cohort relative to that of patients from other African countries may therefore partially be attributed to these therapies.

In the analyses done in this study, active tuberculosis was consistently an independent predictor of death. The risk ratios calculated in this study, before and after adjusting for AIDS-defining illness and CD4+ lymphocyte count, indicate that mortality was consistently higher in cases than in non-cases. HIV-infected patients with active tuberculosis were shown to have a risk of death twice as high as that of HIV-infected patients without active tuberculosis. This higher mortality was more apparent in patients without a history of AIIDS-defining illness and in patients with a CD4+ lymphocyte greater than 200 cells/µl at inclusion. This indicates that tuberculosis increases the risk of death even at the early stages of HIV-infection, and that most of deaths occurred in this cohort at the early stages of HIV infection can be attributed to tuberculosis. This is in agreement with studies done in Africa were TB was shown, in general, to be the primary cause of mortality in HIIV-infected patients, and; more specifically, to be responsible for mortality that occurs at the early stages of HIV infection [8-11]. Previous epidemiologic studies suggest that tuberculosis may accelerate the clinical course of HIIV-infection. In Zaire, the relative risk of death was 2.7 in those with active tuberculosis compared with those without active tuberculosis [8]. A similar estimate for this odds ratio for death was observed in Whalen study

(2.17). These estimates are not different from the estimates of the odds ratio observed in this study (2.37 and 1.70; before and after adjusting for the transient reduction of the CD4 T-lymphocyte induced by active tuberculosis; respectively). This indicates a consistency in both the direction and magnitude in the measure of effect.

It has recently been proposed that tuberculosis may enhance the pathogenecity of HIV infection and accelerate the course of HIV disease. It was discussed in a previous section of this study that the immune activation in response to tuberculosis may play a role in the pathogenesis of HIV-1 infection, by altering the replication of HIIV in target cells, up-regulating HIV expression in T-cells and macrophages, increasing HIIV-1 viral load, and partially suppressing the factors responsible for controlling HIV replication. These factors combined could participate in an increased progression to AIDS and mortality subsequent to the onset of tuberculosis. Thus epidemiologically and biologically plausible that the greater mortality rate that occurred in tuberculosis patients relative to the comparison group in this study can be attributed to active tuberculosis rather than to differences between the two groups in immunosuppression at baseline.

5.1.2 Effect of tuberculosis on incidence of AIDS defining illness

It was observed in this cohort of HIIV-infected patients with active tuberculosis developed AIDS significantly faster than HIV-infected patients without active tuberculosis. This accelerated progression to AIDS after the onset of tuberculosis persisted even after controlling for confounding. HIV-infected patients with active

tuberculosis in this cohort had 60% higher risk of developing AIDS compared with the comparison group of the HIV-infected patients without active tuberculosis.

In addition, active tuberculosis patients had a higher rate of subsequent AIDS-defining illnesses than did the comparison group. Incidence of AIDS-defining illnesses was consistently higher in cases than non-cases of active tuberculosis even after adjusting for CD4+ lymphocyte count, age and antiretroviral therapy. This indicates that HIV patients with active tuberculosis tended to develop AIDS-defining illnesses at a greater rate than non-cases of active tuberculosis over the same period, and after controlling for factors other than active tuberculosis associated with AIDS-defining illnesses. This finding is similar to that observed in the most recent study from the USA. In that prospective cohort study, Munsiff *et al.* observed that tuberculosis was associated with increased risk of development of AIDS-defining illness (RR= 4.1; 95% confidence intervals 1.9-8.7) [12]. Whalen *et al.* observed a higher occurrence, though not significantly different, of AIDS-defining illness among tuberculosis cases compared to a comparison group of non-tuberculosis cases [2]. However, Leroy *et al.* observed no difference in their study and argued that the greater occurrence of AIDS-defining illness in the active tuberculosis patients than in the comparison group observed in Whalen's study might have been due to a detection bias, whereby active tuberculosis patients might have been followed more closely than the patients in the comparison group [2].

It is difficult to attribute the higher rate of subsequent AIDS-defining illness occurring in patients with active tuberculosis in this study to detection bias, since cases of active

tuberculosis were followed in a manner comparable to that of the comparison group [median follow-up = 11.5 *vs* 14.5 months respectively; $p = 0.7$].

However, a history of AIDS-defining illness on its own was predictive of mortality. HIV-infected patients with a history of AIDS-defining illness were estimated to have a risk of death 200% greater than HIV-infected patients who were free from AIDS-defining illness at inclusion.

5.1.3 Effect of tuberculosis on CD4+ T-lymphocyte count

It could be argued that a single measurement of the CD4+ lymphocyte count is insufficient to evaluate the progression of HIV infection, since tuberculosis tends to induce by itself a decline in the CD4+ lymphocyte count [1,2]. The two studies mentioned above reported depressed immunity among HIV-infected patients at the time active tuberculosis was diagnosed. In another study in South Africa, the CD4+ lymphocyte count of a group of HIV-infected patients with tuberculosis was significantly lower than that of a comparison group with active tuberculosis and without HIV infection (median = 230 cells/μL, interquartile range 90-475 *vs* median = 630 cells/μL, interquartile range 500-865 respectively, $p < 0.0001$) [11].

A similar result was observed in this study. The CD4+ lymphocyte count 3-6 months *before* the diagnosis of active tuberculosis was significantly higher than the CD4+ lymphocyte count *at* the diagnosis of active tuberculosis. The CD4+ lymphocyte count 3-6 months *after* the diagnosis of active tuberculosis was also significantly higher than the CD4+ lymphocyte count *at* the diagnosis of active tuberculosis.

However, no significant difference was observed between the median CD4+ lymphocyte counts *before* and *after* the diagnosis of active tuberculosis.

This finding suggests that the reduction in the CD4+ lymphocyte count caused by active tuberculosis was transient. Thus it was important to control for this transient reduction in the CD4+ lymphocyte count at time of active tuberculosis diagnosis in the Cox proportional hazards regression model, to ensure valid estimates for the predictors of survival. Unlike the study in this thesis, none of the previous studies made this adjustment.

When using the CD4+ lymphocyte count *at* date of tuberculosis diagnosis, the adjusted relative risk of death due to active tuberculosis was 2.37, similar to the adjusted risk ratio observed in Whalen's study (2.17), or in the study from Zaire (2.7), but greater than that observed in Leroy's (1.59). A somewhat lower risk ratio was observed after substituting the CD4 count 3-6 months *before* or *after* date of tuberculosis diagnosis in the regression model (risk ratio = 1.70). However, the effect measure remained significant, and the interpretation unchanged.

The significant increase in the CD4+ lymphocyte count *after* the diagnosis of active tuberculosis quantifies the rate of increase in circulating CD4+ T lymphocytes after antituberculosis therapy. The improvement of the median CD4+ lymphocyte count, if sustained, in response to therapy for tuberculosis is likely to have a positive influence on the course of HIV infection. Whalen *et al.*, in recent randomized placebo controlled trial study done in Uganda, argued that the duration of the positive effect of antituberculosis therapy remains to be established especially in areas endemic for

tuberculosis where the high annual risk of infection may affect the risk of tuberculosis after preventive therapy has been completed, especially in persons with immunosuppression [13]. However, the transient reduction in CD4+ lymphocyte count at tuberculosis diagnosis may merely represent migration to the site of tuberculous disease

5.1.4 Other Predictors of mortality

5.1.4.1 Antiretroviral therapy

Antiretroviral therapy was not widely used in this cohort. This may be explained by the fact that most of the patients in this cohort belong to the lower socioeconomic strata of Cape Town and where thus unable to pay for this expensive therapy. As reported in section 4.3.1, only 3 (1.9 %) cases of active tuberculosis and 44 (9.7 %) non-cases were reported to have used antiretroviral therapy. The association between antiretroviral therapy and mortality was consistently non-significant in this study (risk ratio RR = 1.29), in one model, and (risk ratio RR = 1.07) in the model that accounted for the transient reduction in CD4+ lymphocyte induced by active tuberculosis. However, the power of this study to investigate the effect of antiretroviral therapy was low. The limited use of antiretroviral therapy and the relatively small sample of HIV infected patients with active tuberculosis make it difficult to detect any significant association.

Antiretroviral therapy has been shown to prolong survival in patients with AIDS and, in HIV-infected patients without an AIDS-defining condition to delay the progression to AIDS [14-17]. Zidovudine, the main antiretroviral used in this cohort, was the first

antiretroviral agent shown to be of benefit in the treatment of HIV infection. It was shown to decrease transient plasma RNA levels and increase circulating CD4 cells count, decrease the number of AIDS-defining illnesses and progression to AIDS, prolong survival in persons with AIDS, and decrease progression to AIDS in patients with HIV disease [14-17]. However, none of the drugs currently available to treat HIV-infected patients can eradicate infection, although they can decrease the viral load and delay immunologic decline. The reverse transcriptase inhibition associated with nucleoside analogues (e.g., zidovudine) protects uninfected cells, primarily CD4 lymphocytes, from infection [14-17]. However, they have no antiviral effect on cells already infected [14-18]. Antiretroviral therapy has been found in other studies to result in a non-significant improvement in survival. Chaisson *et al.* observed the same result and argued that "the survival advantage conferred by Zidovudine is constrained by time"; and that "although Zidovudine clearly prolongs survival in advanced HIV disease, recent studies have indicated survival is prolonged only for one to two years" [19].

Relatively few HIV infected patients in South Africa can afford antiretroviral therapy. The survival effect of such therapy on HIV-infected South African patients thus has yet to be documented.

5.1.4.2 Co-trimoxazole prophylaxis

In the different analyses done in this study, co-trimoxazole prophylaxis was an independent predictor of improved survival. In the general cohort, use of co-trimoxazole prophylaxis reduced mortality by nearly half in the group using it as a

prophylaxis against AIDS-defining illness, compared to those who did not. The efficacy of co-trimoxazole prophylaxis in reducing mortality and morbidity was specifically examined in the tuberculosis group, and is discussed in a later section of this study.

5.1.4.3 Sociodemographic factors

Sociodemographic factors, such as age, gender, race or ethnic origin and socioeconomic status, may play a role in survival after HIV infection. Although it is not possible to alter most of these factors, their relationship with survival may provide clues to the underlying pathogenesis of infection with HIV. In addition, identification of the relationships between demographic factors and survival is important for predicting individual patients' prognosis.

Age, in some reports, was argued to be an important factor that may confound the association between active tuberculosis and mortality [20-21]. It was observed that older people are more likely to experience shortened survival following HIV infection. The underlying reason for this association, which is also found when considering the time from HIV infection to development of AIDS, is not known. However, Mackall *et al.* have related this association to a poorer capacity for lymphocyte production in older persons [22]. Ben-Yehuda *et al.* have argued further that age may be a marker for some characteristic that makes it more difficult for older people to resist the pathological effect of HIV [23].

In this cohort, however, older age was not significantly predictive of mortality, (risk ratio RR = 1.23, 95% confidence interval = 0.81-1.87, p = 0.3). Age in this cohort ranged from 18 to 69 years [median = 32 years, interquartile range 25-37]. Only 3 patients were older than 65 years, and most of the patients were much younger. This may explain the non-significant effect of older age in survival on this cohort.

To investigate the effect of socioeconomic status on survival in this cohort, patients were classified into two groups. The reference group included patients of higher socioeconomic status as defined by higher employment, education, occupation, coverage by a medical aid schemes, number of dependents, and residence in areas characterized by better living conditions, relative to the comparison group of lower socioeconomic status. Using this classification scheme, it was effectively the same as comparing the different "racial" groups in this cohort, as higher status group included 95.5% of the "white" patients in this cohort, and the comparison group included 97.3% of the "Black" and "Coloured" patients. The median survival of the group of higher socioeconomic status (50 months) was significantly greater than that of the group of lower socioeconomic status (30 months). However, this difference was not observed after controlling for other differences between the groups *viz*. baseline CD4+ lymphocyte count, antiretroviral therapy, co-trimoxazole prophylaxis, age, history of AIDS-defining illness at baseline and active tuberculosis. This result suggests that factors such as background prevalence of tuberculosis, and access to medical care may play an important role in the poorer survival associated with socioeconomic status, or alternatively, "race" among HIV infected patients.

There is controversy over whether race or ethnicity affects survival of HIV-infected patients independently of socioeconomic status. This may be due to the under-representation of the minority ethnic groups in some studies in developed countries. Historically, early studies from the United States that were of sufficient size to consider survival differences between racial groups showed that African American and Hispanic patients had poorer survival than whites after HIV infection [24-25]. This difference in survival may be explained by poorer access to medical care. Minority ethnic groups tend to reside in areas where there is less medical care and treatment. Also their disease may go unnoticed for longer, making it more life threatening and serious when it is finally diagnosed. In contrast, the majority of more recent studies show no survival differences by race or ethnicity [26-27]. A recent study by Chaisson has placed many aspects of the early survival studies into perspective. He has demonstrated that provided patients have equal access to medical care, there is no difference in survival or disease progression. This pertains irrespective of gender, race or socieconomic group. He argued that differences in survival rates are most probably related to availability of medical care and compliance with prophylactic medication, rather than biologic differences in subgroups of patients [19]. Studies of African patients who live in London have suggested no marked differences in survival compared with estimates from developed countries [28]. In common with the studies from Brazil and Thailand [29,30], survival in HIV-infected patients in Africa is generally thought to be poorer as compared with that in developed countries, probably reflecting limitations in access to treatment and care and the high prevalence and incidence and poor control of other infectious diseases, such as tuberculosis.

There are, however, other potential explanations of the observed reduced survival in the active tuberculosis group in this study, reflecting limitations in the study.

It is difficult to determine what proportion of deaths in the case group where directly due to active tuberculosis alone. This is because no information about cause of death was available.

Furthermore, death from tuberculosis may be associated with non-compliance with treatment, or failure to response to antituberculosis therapy particularly if patients are infected with a drug resistant strain of *M. tuberculosis* [31]. No information about compliance was available in this study. It is therefore not possible to exclude treatment non-compliance as a contributor to the observed differences in survival. However, in the general cohort to which the patients of this study belong, Post *et al.* have shown that HIV infection is not associated with an increased rate of drug-resistant tuberculosis [32]. The regimen used for treating tuberculosis in this cohort is the standard therapy to which HIV-infected patients generally respond well.

There were 38 tuberculosis patients who did not meet the inclusion criteria of this study and were thus excluded because either they appeared in the cohort only once or did not avail themselves regularly to follow-up. If these patients experienced a different pattern of survival from that observed in the cases retained in this study, this systematic exclusion may have introduced a biased estimate of the true association between active tuberculosis and mortality. However, it is difficult to determine the magnitude or the direction of this bias.

Referral bias can also not be excluded, since subjects were recruited from teaching hospitals. Such subjects usually present with more advanced disease when they were referred to the HIV clinics for evaluation and proper management. Therefore it is possible that there may be HIV-infected patients with different characteristics (e.g. less immunosuppression or any other characteristics that may affect the prognosis of HIV-infected patients) who did not present to the two hospitals in this study. If the proportion of such patients is significantly large in Cape Town, this clinic bias may account for the poor prognosis observed in this cohort, and may also affect the generalizability of the findings of this study to the general population of adult HIV-infected in Cape Town. However, no other HIV clinics in the public sector in the region were available for HIV-infected patients between 1992 and 1996 other than two HIV clinics in this study.

Viral load is a more specific measure of immunodeprression but was not measured in the present study. Possible differences in viral burden at baseline existed between the two groups of this cohort, may account for the observed differential survival and incidence of AIDS-defining illness. It has been shown that viral load measured by polymerase chain reaction predicts progression of disease even when controlling for the level of CD4+lymphocyte count [33].

5.2 EFFECT OF SITE OF TUBERCULOSIS ON SURVIVAL OF HIV-INFECTED PATIENTS

The reported non-significant difference between the survival of patients with pulmonary and extrapulmonary tuberculosis in this study suggests that patients with extrapulmonary tuberculosis have no worse prognosis than those diagnosed with

pulmonary tuberculosis. In another study done in Cape Town, Post *et al.* observed the same result [34]. Patients with extrapulmonary tuberculosis were observed to have a survival pattern comparable to the survival of other patients without extrapulmonary involvement. Shafer *et al.* observed a similar finding in 54 consecutive HIV-seropositive patients with newly diagnosed tuberculosis and no other AIDS-defining illnesses in the USA [35]. They found no survival difference between patients with pulmonary and extrapulmonary tuberculosis (risk ratio RR = 1.1; 95% confidence interval = 0.4-2.9). However, Whalen *et al.* observed a different result. In their study, also done in the USA, the median survival was shortest in patients with both pulmonary and extrapulmonary disease (8.4 months), followed by extrapulmonary disease alone (15.6 months), while those with pulmonary disease alone survived the longest (30.4 months) ($p < 0.001$) [4].

5.3 EFFECT OF CO-TRIMOXAZOLE PROPHYLAXIS ON MORBIDITY AND MORTALITY OF HIV-INFECTED PATIENTS WITH TUBERCULOSISS

A wide range of severe AIDS-defining illnesses occur in HIV-infected patients with advanced immunodeficiency. Prophylaxis against AIDS-defining illnesses is known to prolong life, decrease morbidity and delay the progression of HIV disease [36]. The success of the prophylaxis against some AIDS-defining illnesses which were reported to be associated with substantial morbidity or mortality [e.g. *Pneumocystis carinii* pneumonia (PCP)] [37] led to a search for effective regimens for the prevention of other AIDS-defining illnesses.

Trimethoprim was specifically developed in the late 1960s as a sulphonamide and was launched in combination with sulfamethxazole as co-trimoxazole (TMP-SMX or

Bactrim®). Co-trimoxazole is a mixture of trimethoprim and sulphamethoxazole in the proportions of 1 part to 5 parts [38]. Co-trimoxazole prophylaxis in HIV-infected patients prevents *Pneumocystis carinii* pneumonia (PCP), a major cause of morbidity and mortality in AIDS patients from developed countries [39-40] This intervention is not widely used in sub-Saharan Africa where PCP is uncommon [39-40]. However, co-trimoxazole also prevents other AIDS-defining illnesses (salmonellosis, isoporiasis, cerebral toxoplasmosis, and other bacterial pneumonia) which occur commonly in HIV-infected patients in Africa [41-42].

Many authors have advocated the use of co-trimoxazole prophylaxis, and documented in their studies an improved prognosis and a decreased incidence of many AIDS-defining diseases among their patients [35-50]. Wiktor *et al.,* in a recent study done in Cote d'Ivoire, have shown that in HIV-infected patients with active tuberculosis, daily co-trimoxazole prophylaxis significantly decreased mortality (46% risk decrease, 95% confidence interval = 23 - 62, $p < 0.001$) and hospital admission rates (43% risk decrease, 95% confidence interval = 10 - 64, $p = 0.02$) [43]. Fischi *et al.* found that only 4 (13%) of 30 patients assigned to co-trimoxazole developed *Pneumocystis carinii* pneumonia (PCP) compared with 16 (53%) of 30 patients who were not receiving prophylaxis ($p < 0.005$) [44]. The four patients who developed *Pneumocystis carinii* pneumonia (PCP) in the co-trimoxazole arm discontinued prophylaxis before the diagnosis because of drug toxicity. Co-trimoxazole prophylaxis was associated with longer median survival (23 months compared with 13 months, $p < 0.002$). Edge *et al.* in a case-control study found a protective benefit of co-trimoxazole on the development of bacteraemia (odds ratio = 0.28; $p = 0.001$) [45]. Furthermore, co-trimoxazole has been found to be effective in preventing cerebral toxoplasmosis. In

one study, none of the 60 patients receiving co-trimoxazole prophylaxis developed toxoplasmosis compared with 12 (13%) of 95 patients of a comparison group who did not receive co-trimoxazole prophylaxis; ($p < 0.05$) [46].

In this study, the efficacy of co-trimoxazole prophylaxis in prolonging survival was firstly examined among the HIV-infected patients in the general cohort when controlling for co-trimoxazole prophylaxis as a probable confounder for the effect of active tuberculosis on mortality, and then specifically in the group of active tuberculosis. The effect of co-trimoxazole prophylaxis in the general cohort was discussed in section [5.1.4.2]. In this section the efficacy of co-trimoxazole prophylaxis in tuberculosis group is discussed.

The tuberculosis group was stratified according to use of co-trimoxazole prophylaxis. Mortality and incidence of AIDS-defining illnesses which could be prevented by co-trimoxazole (PCP, cerebral toxoplasmosis, non-typhoid salmonellosis and bacterial pneumonia) were significantly lower in the co-trimoxazole group compared to the comparison group, and the median survival of the co-trimoxazole group was longer. It is noteworthy that the better outcome in patients given co-trimoxazole was achieved despite their lower CD4+ lymphocyte count at baseline (median 174 cells/μl, interquartile range 33-199, compared to 308 cells/μl, interquartile range 290-745; $p<0.001$).

5.4 RISK FACTORS FOR DEVELOPING TUBERCULOSIS IN HIV-INFECTED PATIENTS

5.4.1 Risk of developing TB

The high cumulative incidence (9%) reported during the 5 years among this cohort can be attributed to the development of the HIV epidemic in a population with one of the highest recorded tuberculosis prevalence rates in the world. The incidence of active tuberculosis in this cohort is considerably greater than that reported in the general population of Cape Town during the period of this study (Table 2-4). This suggests that the HIV epidemic is one of major contributors to the rapidly growing incidence of tuberculosis in Cape Town.

HIV infection confers the greatest known risk for the development of tuberculosis, both for the reactivation of latent infection and for progressive primary disease. Indeed HIV infection is the only factor which has been able to disturb the balance between the tubercle bacillus and man, in the absence of man-made interference. The impact of HIV infection on the epidemiological situation of tuberculosis is so large that, under certain conditions, the tools available at present for tuberculosis control will fail to restrain the increase in the incidence of tuberculosis caused by HIV infection. A strong association exists between tuberculosis and HIV in Africa. The interaction between HIV and tuberculosis fuels the growth of active tuberculosis within the community, thereby enhancing the tuberculosis epidemic. During the last decade, countries to the north of South Africa experienced a dramatic increase in the incidence of tuberculosis [51-53]. Percentage increases compared with the pre-HIV era, have been quoted to be between 250% (Kinshasa and Bangui) and 700% (Lusaka) [53]. However, the current risk of tuberculosis infection and its trend is the most decisive factor in containing the

deterioration of the epidemiologic situation of tuberculosis in developing countries in the future, caused by HIV infection.

In countries with high prevalence of both tuberculosis and HIV infection it is imperative to achieve and maintain a high cure rate of all diagnosed smear positive tuberculosis cases with short-course chemotherapy. Since many tuberculosis cases among HIV-infected persons are smear-negative but culture positive, or smear-negative and culture negative, or culture-positive, it is necessary, whenever possible, to improve case detection of smear-negative cases through screening by X-ray of the chest patients suspected of having tuberculosis, and to examine those with pathology on the X-ray but with negative microscopy, by culture for the tubercle bacilli [54-56].

5.4.2 Effect of advanced immunosuppression on developing overt TB disease

A major finding of this study was that the risk of developing active tuberculosis increased markedly with advancing HIV disease. The analysis which was done to ascertain the factors predictive of active tuberculosis in HIV-infected patients, suggests that HIV-infected patients presenting without evidence of active tuberculosis and with advanced immunnodeficiency, as measured by CD4+ T lymphocyte count (< 200 cells / μL), have a 6-fold increased risk of developing active tuberculosis compared with those with counts > 200 cells / μL.

The analysis suggests further that patients presenting with higher stages of HIV-infection, (WHO stage 3 and 4) or with an AIDS-defining illness are at an increased risk of developing active tuberculosis. AIDS-defining illnesses are markers of a high level of immunosuppression; a level at which the risk of active tuberculosis increases

for patients infected with both HIV and tuberculosis. The significant association between active tuberculosis and the other AIDS-defining illnesses observed in this study may be due to the common underlying immunosuppression. The onset of the AIDS-defining illness leads to a great depletion in CD4+ T-lymphocyte count and thus set the stage for the active tuberculosis to develop.

5.4.3 Effect of race group and socioeconomic status on developing overt TB disease in HIV-infected patients

A higher risk of developing TB was observed among the Black and Coloured HIV-infected patients of this cohort (RR= 3.41) than in White patients . The higher incidence of tuberculosis in these groups is a reflection of the endemicity of tuberculosis in this population group. The racial group variations in the risk of developing TB in this cohort are similar to those reported in other studies done in South Africa [57-58]. This finding indicates that HIV-infected patients with active tuberculosis in Cape Town tend to belong to the less privileged sector of society, and that the risk of developing TB in HIV-infected patients correlates with poverty areas. This fact has implications for access to health services, disease progression and survival.

The basis of the racial difference in tuberculosis has been ascribed to social factors, such as malnutrition, stress, overcrowding, poor access to medical facilities, delay in seeking treatment, (which leads to greater dissemination of the disease), and poverty

[59-61]. However. other studies have suggested that group variation in susceptibility to tuberculosis may be due to genetic differences in the ability of unstimulated macrophages to ingest and lyse the bacilli. In a study done in the USA, Crowle has that there is considerable variation in the vigour with which human macrophages ingest and kill *M. tuberculosis* [62]. He investigated the cellular basis of this putative susceptibility *in vitro* by comparing responses of blood-derived macrophages from Black and White donors to experimental infection with virulent tubercle bacilli. Using microscopic count, the uptake and replication of the tubercle bacilli in these cells in his study were measured at 0,4, and 7 days. He observed that Black-donor phagocytes killed more bacilli during phagocytosis than White-donor phagocytes did. However, the bacilli grew consistently and significantly faster in successfully infected macrophages from Black than from White donors, especially in the presence of Black donor serum ($p < 0.001$). He argued that " these results demonstrate some inherent and environmental liabilities in the monocytic phagocytes and serum of Black people compared with White people, which may contribute to their greater susceptibility to tuberculosis. This appear to furnish an explanation at the cellular level of the difference in infectibility" observed in his study. Stead *et al.* In astudy done in the USA, observed that there were " individual variations in the threshold that an inoculum of *M. tuberculosis* must exceed to establish an infection". He argued that this threshold was significantly higher in the White than in the Black patients in his study [63]. However, the generalizability of these results to the universal "racial' differences remains controversial.

The prevalence of past *M. tuberculosis* infection is known to be higher in older people. The lack of association between older age and tuberculosis in this study could

be explained by the fact that the majority of the patients in this cohort were of young age.

It was difficult to ascertain whether the high percentage of unemployed patients in this cohort was a consequence of their HIV status. Even without HIV infection, unemployment in many Western Cape suburbs easily exceeds 50%. However, HIV infected people are less likely to maintain their occupational status than seronegative persons, as they are more likely to lose their jobs and become financially dependent upon family and friends. In the cohort of this study where unemployment is high among HIV-infected patients, a household of several adults and children was frequently dependent on the earnings of a single member. That person is usually the young adult who is also sexually active, unmarried, uneducated and most at risk of acquiring HIV.

Higher level of education was observed to be protective against the risk of active tuberculosis. Higher education in this study is likely to be a marker of higher socioeconomic status which is a known protective factor against developing active tuberculosis. Lack of education has been reported previously to be associated with risk of developing active tuberculosis and poorer survival in patients with and without HIV-infection in African countries [64].

While previous studies in Africa have shown a strong association between HIV infection and the risk of tuberculosis, they have not demonstrated a marked increase in tuberculosis incidence with HIV disease progression. Two studies from Central and West Africa have reported higher CD4+ T-lymphocyte count of patients with

tuberculosis and HIV-infection than in the present study [63,65]. Differing methodology for tuberculosis diagnosis may partially explain the apparent conflicting findings. A cross-sectional study in Zaire [65] reported that pulmonary tuberculosis occurred across the spectrum of HIV disease, with the majority of tuberculosis (67%) associated with CD4+ T-lymphocyte count above 200/μL. The study was limited to sputum smear positive pulmonary disease, which may have under-diagnosed the more atypical presentations associated with advanced immune suppression [67]. A study in Côte d'Ivoire of smear positive pulmonary TB and clinically diagnosed extrapulmonary tuberculosis, reported 57% of cases occurred at CD4+ T-lymphocyte count above 200/μL [66]. In contrast, post mortems from Zaire and Côte d'Ivoire showed histological and culture evidence of disseminated tuberculosis in 41% and 54% of AIDS patients respectively [42,68] with a median CD4+ T-lymphocyte count of 88/μL in the Zaire study [42]. The facilities for histology and tuberculosis culture available at the 2 hospitals in the present study may have increased case finding, as radiographic presentation of pulmonary tuberculosis in the HIV-infected patients in the general cohort to which the patients in this study belong is frequently atypical and 52% are sputum smear negative [67]. A recent prospective multicentre USA cohort study found the risk of developing TB was significantly higher in patients with a CD4 CD4+ T-lymphocyte count <200/μL [69].

The cellular mechanisms underlying the development of tuberculosis in HIV-infected patients have been discussed in a previous section. In brief, inhaled tubercle bacilli enter the lungs as droplet nuclei where they are engulfed by alveolar macrophages which process the mycobacterial antigens and resent them to CD4+ T helper lymphocytes. In healthy individuals, infection with *M. tuberculosis* is probably

controlled by a dynamic interaction between CD4+ helper T lymphocytes and tissue macrophages containing intracellular tubercle bacilli, resulting in granuloma formation. Macrophages are activated to kill intracellular organisms by lymphokines, but whereas most intracellular infections can be inhibited or killed by interferon γ induced activation [70], *M tuberculosis* in human macrophages systems seems relatively resistant suggesting that other mechanisms are involved [71]. Forte *et al.* suggest that a class of CD4+ cytotoxic T- lymphocyte is involved in the control of *M tuberculosis* latent infections by causing cytolysis of infected macrophages, thus releasing tubercle bacilli and exposing them to further immune attack [72]. HIV disturbs this process by infecting macrophages and upsetting their function [73]. Thus, HIV infection is associated with a steady decrease in both numbers and activity of CD4+ lymphocytes [74]. In conclusion, the findings of this study confirm that HIV-infected patients are at higher risk of developing active tuberculous disease.

5.4.4 Prophylaxis and treatment of TB in HIV-infected patients

Despite the higher risk of tuberculosis described above, there is evidence from several sources that tuberculosis may respond well to treatment, even in the presence of HIV infection. Furthermore, the efficacy of isoniazid (INH) prophylaxis has been demonstrated in various studies. Pozniak argued that isoniazid is "the most potent bactericidal drug and kills more than 90% of bacilli within 7 days by acting on metabolocally active bacilli, and effective at preventing the emergence of drug resistance"[7]. Pape *et al.* found that the administration of INH for one year in Haitian patients reduced the frequency of tuberculosis from 10 to 1.7/100 person-years [75]. In addition, INH was shown to reduce the risk of tuberculosis in persons with HIV

infection in Zambia, Uganda, Mexico and the United States [76]. In all these countries, the protective effect of INH was greater when the incidence of tuberculosis was high than when it was low.

However, while antituberculosis prophylaxis is believed to eradicate the HIV-infected patient's bacilli and therefore prevent reactivation tuberculosis, this is unlikely to subsequently protect from a new tuberculosis infection several years later [75]. This necessitates constant screening for tuberculosis in HIV-infected patients even after successful treatment.

5.5 VALIDITY OF HIV STAGING SYSTEMS IN HIGH TB PREVALENCE SETTING

In this cohort, a median survival of 24 months was observed following the diagnosis of active tuberculosis. Furthermore, it was shown that the prognosis of this group was significantly different from that of patients diagnosed with an AIDS-defining illness. These findings suggest that the diagnosis of active tuberculosis in HIV-infected patients is not necessarily associated with a prognosis similar to that of other AIDS-defining illnesses.

Using the WHO and CDC staging systems, survival of active tuberculosis patients was comparable to WHO stage 3 and to CDC stage B. More specifically, the prognosis of HIV-infected patients with active tuberculosis was equivalent to that of HIV-infected patients with oral hairy leukoplakia or oral candidiasis.

More than a decade has now elapsed since the initial description of AIDS and much more information is now available about the natural history of infection with HIV. Observations from a number of prospective studies have provided insight into the characteristic changes in laboratory parameters (most notably CD4 lymphocyte count and appearance of symptoms) that are markers of progression of HIV disease [78-81]. Since the beginning of the epidemic, a number of classifications have been proposed to stage HIV infected individuals throughout the course of their disease. The clinical case definition for AIDS in adults was developed at a WHO workshop on AIDS in Bangui, Central African Republic, in October 1985 [82], and later revised [83]. Staging systems play a critical role in management of HIV infected patients because they assist clinicians in selecting appropriate prognostic stratification for clinical studies, and identify intermediate measures of disease progression that can be used to expedite the assessment of potentially promising therapies. The effectiveness of an HIV staging system will depend on consistent prognostic utility despite heterogeneous populations. As has been discussed in a previous section of this thesis, a major limitation of the existing staging systems is that the clinical case definitions lack specificity in distinguishing cases of tuberculosis from cases of AIDS. Some other disadvantages of these systems are that they are not necessarily based on severity of illness, and a large number of people cannot be classified. These staging system often place all patients with AIDS into one group, whereas the heterogeneity in survival of patients with AIDS suggests that a finer distinction might be more useful. Furthermore, differences in the spectrum of disease and in the population may significantly decrease the specificity of these staging systems, e.g. in a high endemic tuberculosis setting.

Few studies, however, have specifically addressed the ranking of AIDS-defining diseases and survival after each diagnosis [82-83]. A natural order of AIDS-defining diseases, based on the average CD4 lymphocyte count at which they occur, has been suggested [78-81,85-86]. This order ranges from diseases such as lymphomas, tuberculosis and Kaposi's sarcoma, which typically occur at higher CD4 lymphocyte counts, to diseases such as cytomegalovirus (CMV), which are frequently observed in patients with CD4+ T-lymphocyte count below 50 cells/μl [85].

In many African and other developing countries, laboratory facilities are insufficient to allow reliable diagnosis of the opportunistic infections and malignant disorders required to meet the definition. Strict adherence to the CDC HIV staging systems in African countries would lead to greater-under recognition of the disease. Furthermore, the clinical spectrum of AIDS in Africa may differ substantially from that described in Europe and North America. Testing for HIV antibodies by currently available technology may not be feasible.

The findings of this study may contribute to clarify why in a high tuberculosis prevalence setting and in absence of co-infection with an AIDS-defining illness active tuberculosis may not qualify as an AIDS-defining illness.

These findings suggests further that the CDC criteria (Appendix I) for staging HIV-infected patients is of a limited utility in predicting the course of HIV progression in a high tuberculosis prevalence setting. The CDC staging system may be useful in the setting for which it was initially proposed and validated, but its applicability in other settings is controversial. The universal use of the CDC system has been compromised

by its limited prognostic utility in the past, and a lack of international consensus concerning recent revisions to the original system.

Conclusion and recommendations

Tuberculosis is one of the most important infections affecting HIV-infected patients in the world. A strong interaction between tuberculosis and HIV has been shown in this study and in other seroprevalence surveys in places as diverse as Burundi, the United States, France, Italy, Tanzania, and Haiti. Based on the findings of this study, some observations need emphasis.

- The high prevalence of tuberculosis in the Western Cape makes it mandatory to screen every HIV-patient for tuberculosis. Suppression of active disease by anti-tuberculosis chemotherapy in HIV-infected patients with active tuberculosis should be started immediately following the diagnosis of tuberculosis.

- On the other hand, all adults presenting with tuberculosis need to be assessed for HIV infection because the medical management of tuberculosis may needed to be altered in the presence of HIV infection. Problems arise both when HIV-infected patients are contemplating starting antiretroviral therapy and when they are already on such therapy and tuberculosis is diagnosed. Some guidelines regarding the treatment of tuberculosis and HIV already exist and usually require modification of either tuberculosis therapy or the antiretroviral drugs regimen [87]. Major drug-drug interactions can occur especially between the rifampicin and protease inhibitors and non-nucleoside reverse transcriptase inhibitors

(NNRTI) [87]. In addition, tuberculosis patients who are infected with HIV will benefit by being monitored for early diagnosis of AIDS-defining illnesses and other manifestations of HIV infection.

- Based on the findings of this study, active tuberculosis, in the absence of co-infection with other AIDS-defining illnesses, must not be considered as an AIDS-defining illness. Further studies are needed to replicate the findings of this study, and to evaluate the clinical case definition of AIDS in areas where tuberculosis is highly endemic.

- The findings of this together with those of Wiktor *et al.*[43] indicate a dramatic benefit of prophylactic co-trimoxazole against a variety of AIDS related illnesses in HIV-infected patients with active tuberculosis and confirm that the lower dose of 480 mg daily is also effective in an African context. Co-trimoxazole is an affordable intervention which would help improve the therapeutic nihilism which surrounds HIV in many African countries.

- Many patients in this study presented at late stage and were severely immunocompromised when their HIV status was initially diagnosed. This deprived them of the opportunity of receiving interventions aimed at improving their prognosis. This may be due to a lack of awareness or knowledge of HIV/AIDS in the areas from which the subjects of this cohort were drawn. Delay in seeking medical attention can be viewed as integral to the poor survival observed in this study. This clearly indicates the need to educate the groups at high risk for HIV with regard to the importance of early diagnosis, and the

monitoring and suppression of HIV infection. However there are many practical difficulties in screening for HIV in its early stages.

- This study indicates that both *Mycobacterium tuberculosis* and HIV have their impact on certain categories of the Western Cape population. The most affected are young adults at the beginning, or in the prime, of their productive lives. This has serious implications for the burden of both diseases in the affected communities. Furthermore, students were reported in this study to be the third highest category affected by HIV infection. It is vital to ensure that young people understand the nature of the AIDS epidemic and the specific actions they can take to prevent HIV infection.

- Antiretroviral therapy is expensive and generally unaffordable for the vast majority of HIV-infected patients in South Africa. At present, few HIV-infected patients are able to afford combination anti-retroviral therapy. Providing these drugs to those who cannot afford to pay for them should be viewed as a priority of the health authorities in South Africa. However the cost implications are great, and this will remain a major barrier in South Africa.

- Co-infection may make the diagnosis of tuberculosis extremely difficult in some HIV-infected patients. The absence of cavitation and the scarcity of tubercule bacilli in the sputum of patients with advanced immunosuppression, may lead to disastrous exclusion of the diagnosis. Failure to diagnose and manage tuberculosis appropriately can result in reduced survival and death of the patient and infection of contacts, including other patients and health-care personnel. To establish the

diagnosis, a variety of specimens, including respiratory secretions, bronchial washings, gastric lavage, lung tissue, pleural fluid, lymph node tissue, bone marrow, blood, urine, stool, brain biopsy, and cerebrospinal fluid, may need to be obtained for mycobacterial culture. Specimens must be examined microscopically, but the inability to demonstrate acid-fast bacilli and the absence of granuloma formation does not exclude the diagnosis of tuberculosis [54-56]. This is particularly relevant to South Africa, were laboratory screening of sputum (without subsequent culture) for AFBs may jeopardise a more aggressive pursuit of mycobacterial infection. However, diagnosing tuberculosis by culture only is both expensive and slow. This clearly indicates the need for developing evidence-based algorithm for clinical diagnosis of smear negative tuberculosis in HIV-infected patients.

- In conclusion, access to decent health care must be viewed as a priority for all South Africa's diverse people. Determination of HIV status, the exclusion of tuberculosis and the provision of a relatively cheap drug such as INH and co-trimoxazole can be all successfully integrated at the primary care level and are suited to the current proposals for health care reform in this country. Should improved control not occur within the next few years, the prevalence of HIV in the adult population will rise to 17-30%. Every effort should be made to undo the cycle of HIV and tuberculosis and reduce their devastating impact on South African society.

REFERENCES

1. Leroy V, Salmi R, Dupon M, et al: **Progression of human immunodeficiency virus in patients with tuberculosis disease.** *Am J Epidemiol* 1997; 145:293-300.
2. Whalen C, Horsburgh CR, Hom D, et al. **Accelerated course of human immunodeficiency virus after tuberculosis.** *Am J Respir Crit Care Med* 1995; 151: 129-135.
3. Stoneburner R, Laroche E, Prevots R, et al. **Survival in a cohort of Human Immunodeficiency Virus-infected tuberculosis patients in New York city.** *Arch Intern Med* 1992; 152:,2033-2037.
4. Whalen C, Horsburgh CR, Hom D, et al. **Site of disease and opportunistic infection predict survival in HIV-associated tuberculosis.** *AIDS* 1997, 11: 455-460.
5. Perneger TV, Sudre P, Lundgren JD, Hirschel B. **Does the onset of tuberculosis predict shorter survival. Results of a cohort study in 17 European countries over 13 years.** *Br Med J* 1995; 311:1468-1471.
6. Leroy V, Msellati P, Salmi R, et al. **Four years of natural history of HIV-1-infection in African women: a prospective cohort study in Kigali (Rwanda), 1988-93.** *J Acqir Immune Defic Syndr Hum Retrovirol*1995; 9: 415-421.
7. Pozniak AL, Miller R, Ormerod L. **The treatment of tuberculosis in HIV-infected persons.** *AIDS* 1999; 13: 435-445.
8. Braun MM, Nsanga B, Ryder RW, et al. **A retrospective cohort study of the risk of tuberculosis among women of childbearing age with HIV infection in Zaire.** *Am Rev Respir Dis* 1991; 143: 501-504.
9. Perriens JH, Colebumers RL, Karahunga C, et al. **Increased mortality and tuberculosis treatment failure rate among human immunodeficiency virus**

(HIV) seropositive compared with negative patients with pulmonary tuberculosis treated with "standard" chemotherapy in Kinshasa, Zaire. *Am Rev Respir Dis* 1991; 144: 750-755.

10. Nunn P, Brindle R, Carpenter L, et al. **Cohort study of human immunodeficiency virus infection in patients with tuberculosis in Nairobi, Kenya: analysis of early (6-month) mortality**. *Am Rev Respir Dis* 1992; 146: 849-854.
11. Martin DJ, Sim JG, Sole GJ, et al. **CD4+ T-lymphocyte count in African patients co-infected with HIV and tuberculosis.** *J Acquir Imuune Defic Syndr Hum Retrovirol* 1995; 8: 386-391.
12. Munsiff SS, Alpert PL, Gourevitch MN, chang CJ, Klein RS. **A prospective study of tuberculosis and HIV disease progression**. *J Acquir Immune Defic Syndr Hum Retrovirol* 1998; 19: 361-366.
13. Whalen C, Johnson JL, Okera A, et al. **A trial of three regimens to prevent tuberculosis in Ugandan adults infected with the human immunodeficiency virus**. *New Engl J Med* 1997; 337: 801-808.
14. McLeod GX, Hammer SM. **Zidovudine : Five years later**. *Ann Intern Med* 1992; 117:487.
15. Fischl MA, Richman DD, Hansen N, et al. **The safety and efficacy of zidovudine (AZT) in the treatment of subjects with mildly symptomatic human immunodeficiency virus type 1 (HIV) infections**. *Ann Intern Med* 1990; 112: 727-731.
16. Graham NMH, Zeger SL, Park PL, et al. **The effects on survival of early treatment of human immunodeficiency virus infection**. *N Engl J Med* 1992; 326: 1037.

17. Kinloch-DeLoes S, Hirschel BJ, Hoen B, et al. **A controlled trial of zidovudine in primary human immunodeficiency virus infection**. *N Engl J Med* 1995; 333: 408-413.

18. Swason CE, Tindall B, Cooper DA, for the Australian Zidovudine Study Group. **Efficacy of zidovudine treatment in homosexual men with AIDS-related complex: factors influencing development of AIDS, survival and drug intolerance**. *AIDS* 1994; 8: 625-634.

19. Chaisson RE, Keruly JC, Moore RD, et al. **Race, sex, drug use, and progression of human immunodeficiency virus disease**. *N Engl J Med* 1995; 333: 751-756.

20. Jacobson LP, Kirby AJ, Polk S, et al. **Changes in survival after acquired immunodeficiency syndrome (AIDS): 1984-1991**. *Am J Epidemiol* 1993;138: 952-964.

21. Osmond D, Charlebois, Lang W, Shiboski S, Moss A. **Changes in AIDS survival time in two San Francisco cohorts of homosexual men, 1983-1993**. *JAMA* 1994, 271: 1083-1087.

22. Mackall CL, Fleisher TA, Brown MR, et al. **Age, thymopoiesis, and CD4+ T-lymphocyte regeneration after intensive chemotherapy**. *N Engl J Med* 1995; 332: 143-149.

23. Ben-Yehuda A, Weksler ME. **Immune senescence : mechanisms and clinical implications**. *Cancer Invest* 1992; 10: 525-531.

24. Blum S, Singh TP, Gibbons J, et al. **Trends in survival among persons with acquired immunodeficiency syndrome in New York city. The experience of the first decade of the epidemic**. *Am J Epidemiol* 1994; 139: 351-361.

25. Piette J, Mor V, Fleishman L. **Patterns of survival with AIDS in the United States**. *Health Serv Research* 1991; 26: 75-95.

26. Chang HH, Morse DL, Noonan C, et al. **Survival and mortality patterns of an acquired immunodeficiency syndrome (AIDS) cohort in New York state.** *Am J Epidemiol* 1993,138: 341-349.

27. Seage GR, Oddleifson S, Carr E, et al. **Survival with AIDS in Massachusetts 1979-1989.** *Am J Pub Health* 1993; 83: 72-78.

28. Low N, Paine K, Clark R, Mahalingam M, Pozniak AL. **AIDS survival and progression in Black Africans living in South London, 1986-1994.** *Genitoriun Med* 1996; 72: 12-16.

29. Chequer P, Herst N, Hudes ES, et al. **Determinants of survival in adult Brazilian AIDS patients, 1982-1989.** *AIDS* 1992; 6: 483-487.

30. Kitayaporn D, Tansuphaswadikul S, Lohsomboon P, et al. **Survival of AIDS patients in the emerging epidemic in Bangkok, Thailand.** *J Acquir Immune Defic Syndr* 1996; 11: 77-82.

31. Fischl MA, Daikos GL, Uttamchandani RB, et al. **Clinical presentation and outcome of patients with HIV infection and tuberculosis caused by multi-drug-resistant bacilli.** *Ann Intern Med* 1992; 117: 184-190.

32. Post F, Wood R. **HIV infection is not associated with an increased rate of drug-resistent tuberculosis.** *S Afr Med J* 1997; 87: 903-906.

33. Schnittman SM, Greenhouse JJ, Psallidopoulos MC, et al. **Increasing viral load burden in CD4+ T cells from patients with human immunodeficiency virus (HIV) infection reflects rapidly progressive immunosuppression and clinical disease.** *Ann Intern Med* 1989; 113: 438-443.

34. Post FA, Wood R, Maartens G: **Acquired immunodeficiency syndrome in Africa: survival according to AIDS defining illness** *[unpublished study].*

35. Shafer RW, Bloch AB, Vasudavan V, et al. **Predictors of survival in HIV-infected tuberculosis patients.** *AIDS* 1996; 10: 269-272.

36. Gallant JE, Moore RD, Chaisson RE. **Prophylaxis for opportunistic infections in patients with HIV infection.** *Ann Intern Med* 1994; 120: 932-944

37. Feedberg KA, Tosteson AN, Cohen CJ, Cotton DJ. **Primary prophylaxis for *pneumocystis carinii* pneumonia in HIV-infected people with CD4 counts below 200/mm^3 : a cost-effectiveness analysis.** *J Acquir Immune Defic Syndr* 1991; 4: 521-531.

38. British Medical Association, Royal Pharmaceutical Society of Great Britain. **British National Formulary** 1994; No 27.

39. Kovacs JA, Masur H. **Prophylaxis for *pneumocystis carinii* pneumonia in patients with human immunodeficiency virus.** *Clin Infect Dis* 1992; 14: 1005-1009.

40. Phair J, Munoz A, Detels R, Kaslow R, Rinaldo C, Saah A. **The risk of *pneumocystis carinii* pneumonia among men infected with human immunodeficiency virus type I. Multicentre AIDS Cohort study Group.** N *Engl J Med* 1990; 322: 161-165.

41. Grant AD, Djomand G, De Cock KM. **Natural history and spectrum of disease in adults with HIV/AIDS in Africa.** *AIDS* 1997; 11(suppl B): S43-54.

42. Lucas SB, Hounnou A, Peacock C, et al. **The mortality and pathology of HIV infection in a West African city.** *AIDS* 1993; 7: 1569-1579.

43. Wiktor SZ, Morokro MS, Grant AD, et al. **Efficacy of trimethoprim-sulphamethoxazole prophylaxis to decrease morbidity and mortality in HIV-1-infected patients with tuberculosis in Abidjan, Cote d'Ivoire: a randomised controlled trial.** *Lancet* 1999; 353: 1469-1475.

44. Fischi MA, Dickenson GN, La Voie L. **Safety and efficacy of sulfamethoxazole and trimethoprim chemoprophylaxis for *pneumocystis carinii* pneumonia in AIDS.** *JAMA* 1988: 259: 1185-1189.

45. Edge MD, Rimland D. **Community-aquired bacteremia in HIV-positive patients: protective benefit of co-trimoxazole.** *AIDS* 1996 ; 10: 1635-9

46. Carr A, Tindall B, Brew BJ, et al. **Low-dose trimethoprime-sulfamethoxazole prophylaxis for toxoplasmosis encephalitis in patients with AIDS.** *Ann Intern Med* 1992; 117: 106-111.

47. Michele C, Raffi F, Besnier JM, et al. **Cotrimoxazole for primary prophylaxis of toxoplasmosis encephalitis in advanced HIV patients** *[Abstract WS-B13-2]. IX International Conference on AIDS, Berlin: Der Congress* 1993; 1:56.

48. Thea DM, Lambert G, Weedon J, et al. **Benefits of primary prophylaxis before 6 months of age in reducing the incidence of related *pneumocystis carrinii* pneumonia and early death in a cohort of 112 human immunodeficiency virus-infected infants. New York City peri-natal HIV Transmission Collaborative Group.** *Paediatrics* 1996; 97: 59-64.

49. Ruskin J, La Riviere M. **Low-dose co-triomoxazole for prevention of *Pneumocystis carrinii* pneumonia in human immunodeficiency virus disease.** *Lancet* 1991; 340: 1099.

50. Ioannidis JP, Cappelleri JC, Skolnik PR, Lau J, Sacks HS. **A meta-analysis of the relative efficacy and toxicity of *Pneumocystis carinii* prophylactic regimens.** *Arch Intern Med* 1996; 22; 156:177-188.

51. Goodgame RW. AIDS in Uganda-clinical and social features. N Engl J Med 1990; 323: 383-389.

52. Standheart B. **The association of tuberculosis and HIV infection in Burundi.** *AIDS Res Hum Retrovirol* 19989; 5: 247-251.

53. Perriens JH. **Tuberculosis and HIV infection: implications for Africa.** *AIDS* 1991; 5 (suppl 1): S127-S133.

54. Wilkinson D, Moore DAJ. **HIV-related tuberculosis in South Africa- clinical features and outcomes.** *S Afr Med J* 1996: 60-63.

55. Harries AD. **Tuberculosis and human immunodeficiency virus infection in developing countries.** *Lancet* 1990; 335: 387-390.

56. Tshibwabwa-Tumba E, Mwinga A, Pobee JO, Zumla. **Radiological feature of pulmonary tuberculosis in 963 HIV-infected adults at three Central African Hospitals.** *Clin Radiol* 1997; 52(11): 837-41.

57. Wood, Maartens G, O'Keefe E. **The changing pattern of transmission and vlinical presentation of HIV in Western Cape of South Africa (1934-1995).** *S Afr J Epidemiol Infect* 1996; 11: 96-98.

58. Doyle PR. **The impact of AIDS on South Africa population. AIDS in South Africa: the Demographic and economic Implications.** *Johannesberg: Center for Health Policy, University of the Witwatersand* 1991.

59. Bates JH. **Tuberculosis: Susceptibility and resistance.** *Am Rev Respir Dis* 1982; 125: 20-24.

60. Ochs CW. **The epidemiology of tuberculosis.** *JAMA* 1962; 179:247-252.

61. Houk VN, Baker JH, Sorensen K, Kent DC. **The epidemiology of tuberculosis: infection in a closed environment.** *Arch Environ Health* 1968; 16: 26-35.

62. Crowle AJ. **The tubercle bacillus-human macrophage relationship studied in vitro. Mycobacteium tuberculosis: interactions with the immune system.** *Wiley Pub, New York, Wiley and interscience* 1988.

63. Stead W, Senner JW, Reddick WT, Lofgren JP. **Racial differences in susceptibility to infection by *Mycobacterium tuberculosis*.** *N Engl J Med* 1990; 322: 422-427.

64. Nunn P, Bridle R, Carpenter L, et al. **Cohort study of Human Immunodeficiency Virus inpatients with tuberculosis in Nairobi, Kenya.** *Am Rev Respir Dis* 1992; 148; 849-854.

65. Mukadi Y, Perriens JH, St Louis ME, et al. **Spectrum of immunodeficiency in HIV-1-infectedpatients with pulmonary tuberculosis in Zaire.** *Lancet* 1993; 342: 143-146.

66. Achah AN, Coulibay D, Digbeu H, et al. **Response to treatment, mortality, and CD4 lymphocyte count in HIV-infected persons with tuberculosis in Abidjan, Cote d'Ivoire.** *Lancet* 1995; 345: 607-610.

67. Post FA, Wood R, Pillay GP. **Pulmonary tuberculosis in HIV infection: radiologic appearance is related to CD4+ T-lymphocyte count.** *Tubercle Lung Dis* 1995; 76: 517-520.

68. Nelson Am, Perriens JH, Kapita B et al. **A clinical and pathological comparison of the WHO and CDC case definitions for AIDS in Kinshasa, Zaire: is passive surveillance of value?** *AIDS* 1993; 7: 1241-1246.

69. Markowitz N, Hansen NI, Hopewell PC, et al. **Incidence of tuberculosis in the United States among HIV-infected persons.** *Ann Intern Med* 1997; 126: 123-132.

70. Murray HW, Hillman JK, Robin BY, et al. **Patients at risk for AIDS-related opportunistic infections.** *N Engl J Med* 1985; 313: 1504-1510.

71. Rook GAW, Steele J, Ainsworth M, Champion BR. **Activation of macrophages to inhibit proliferation of Mycobacterium tuberculosis: comparison of the**

effects of recombitant gamma-interferon on human monocytes and murine peritoneal macrophages. *Immunology* 1986; 59: 333-338.

72. Forte M, Maartens G, Rahelu M, et al. **Cytolytic T cell activity against mycobacterial antigens in HIV infection.** *AIDS* 1992; 6: 407-411.
73. Fauci AS. **Immunopathogenic mechanisms in human immunodeficiency virus (HIV) infection.** *Ann Intern Med* 199; 114: 678-693.
74. Pantaelo G, Graziosi C, Fauci A. **The immunopathogenesis of human immunodeficiency virus infection.** *N Engl J Med* 1993; 328: 327-335.
75. Pape JW, Jean S, Ho JL, Hafner A, Johnson WD. **Effect of isoniazid prophylaxis on incidence of active tuberculosis and progression of HIV infection.** *Lancet* 1993; 342: 268-272.
76. Bucher HC, Griffith LE, Guyatt GH, et al. **Isoniazid prophylaxis for tuberculosis in HIV infection: a meta-analysis of randomized controlled trials.** *AIDS* 1999; 13: 501-507.
77. Kaplan JE, Masur H, Holmes KK, et al., and the USPHS/IDSA prevention of opportunistic infections working group. **USPHS/IDSA guidelines for the prevention of opportunistic infections in persons infected with human immunodeficiency virus: introduction.** *Clin Infect Dis* 1995; 21(suppl 1.1):S1.
78. Germain RN. **Antigen processing and CD4+ T cell depletion in AIDS.** *Cell* 1988; 54: 441-444.
79. Detels R, English P, Giorgi JV, et al. **Patterns of CD4+ cell changes after HIV-1 infection indicate the existence of a co-determinant of AIDS.** *J Acquir Immune Defic Syndr* 1988; 1: 390-395.

80. Lang E, Perkins H, Anderson RE, etal. **Patterns of T lymphocyte changes with human immunodeficiency virus infection: from seroconversion to the development of AIDS.** *J Acquir Immune Defic Syndr* 1989; 2: 63-69.

81. Stein DS, Korvick JA, Vermund SH. **CD4+ lymphocyte cell enumeration for prediction of clinical course of human immunodeficiency virus disease: A review.** *J Infect Dis* 1992; 165: 352-362.

82. World Health Organisation. **Acquired immunodeficiency syndrome (AIDS).** *Weekly Epidemiol Rec* 1986; 61: 69-73.

83. The WHO International Collaborating group for the Study of the WHO staging system. **Proposed "World Health Organisation Staging system for HIV infection and disease": preliminary testing by an international collaborative cross-sectional study.** *AIDS* 1993; 7: 711-718.

84. Lou K, Law M, Kaldor JM, McDonald AM, Cooper DA. **The role of initial AIDS-defining illnesses in survival following AIDS.** *AIDS* 1995; 9: 57-63.

85. Crowe SM, Carlin JB, Stewart KI, Lucas CR, Hoy JF. **Predictive value of CD4 lymphocyte numbers for the development of opportunistic infections and malignancies in HIV-infected persons.** *J Acquir Immun Defic Syndr* 1991; 4: 770-776.

86. Hanson DL, Chu SY, Farizo KM, John WW. **Distribution of CD4+ T-lymphocytes at diagnosis of acquired immunodeficiency syndrome-defining and other human immunodeficiency virus related illnesses.** *Arch Intern Med* 1995; 155: 1537-1542.

87. Centres for Disease Control and Prevention: **Clinical update: impact of HIV protease inhibitors on the treatment of HIV-infected patients with tuberculosis.** *MMWR* 1996, 45:921-925.

APPENDICES

APPENDIX I

1993 Revised Classification System for HIV Infection and Expanded Surveillance Case Definition for AIDS Among Adolescents and Adults.

The revised CDC classification for HIV-infected adolescents and adults categorizes persons on the basis of clinical conditions associated with HIV infection and CD4+ T-lymphocyte counts. The system is based on three ranges of CD4+ T-lymphocyte counts and three clinical categories and is represented by a matrix of nine mutually exclusive categories. This system replaces the classification system published in 1986, which included only clinical disease criteria and which was developed before the widespread use of CD4+ T-cell testing.

CD4+ T-lymphocyte Categories

The three CD4+ T-lymphocyte categories are defined as follows:

* Category 1 : greater than or equal to 500 cells/ℓl
* Category 2 : 200-499 cells/ℓl
* category 3 : less than 200 cells/ℓl

Clinical Categories:

The clinical categories of HIV infection are defined as follows:

Category A

- Asymptomatic HIV infection
- Persistent generalized lymphadenopathy
- Acute (primary) HIV infection with accompanying illness or history of acute HIV infection

Category B

- Bacillary angiomatosis
- Candidiasis, oropharungeal (trush)
- Candidiasis, vulvovaginal, persistent, frequent, or poorly responsive to therapy
- Cervical dysplasia (moderate or severe)/ cervical carcinoma in situ
- Consistutional symptoms, such as fever (35.5 C) or diarrhoea lasting > 1 month
- Hairy leukoplakia, oral
- Herpes zoster (shingles)
- Idiopathic thrombocytopenic purpura
- Listeriosis
- Pelvic inflammatory disease
- Peripheral neuropathy

Category C (AIDS)

- Candidiasis of bronchi, trachea, or lungs
- Candidiasis, oesophageal
- Cervical cancer, invasive
- Cryptococcosis, extrapulmonary
- Cryptococcosis, chronic intestinal (>1 month)
- Cytomegalovirus disease (other than liver, spleea, or nodes)
- Cytomegalovirus retinitis (with loss of vision)
- Encephalopathy, HIV-related
- Herpes simplex
- Histoplasmosis, extrapulmonary

- Isosporiasis, chronic intestinal (>1 month)
- Kaposi,s sarcoma
- Lymphoma, Burkitt's (or equevalent term)
- Lymphoma, imunoblastic (or equivalent)
- Lymphoma, primary, of brain
- Mycobacterium avium complex or M. Kansasii, dissiminated or extrapulmonary
- Mycobacterium tuberculosis, any site (pulmonary or extrapulmonary)
- Mycobacterium, other speices or unidentified speices, disseminated or extrapulmonary
- Pneumocystis carinii pneumonia
- Pneumonia, recurrent
- Progressive multifocal leukopencephalopathy
- Salmonella septivaemia, recurrent
- Toxoplasmosis of brain
- Wasting syndrome due to HIV

APPENDIX II

World Health Organization Clinical Staging System

I Laboratory Staging

	CD4+ count cells/mm^3	Total lymphocyte count cells$x10^6$/L
A.	>500	>2000
B	200-500	1000-2000
C	<200	<1000

2 Clinical Stages

Stage 1

- Acute retroviral infection
- Asymptomatic HIV infection
- Persistent generalised lymphadenopathy
- Performance scale 1: asymptomatic, normal activity

Stage 2

- Weight loss, less than 10% of normal body mass
- Minor mucocutaneous manifestation (seborrhoeic dermatitis, purigo, fungal nail infections, recurrent oral ulcerations, and angular chelitis.
- Herpes zoster (shingles) within the last five year .
- Recurrent upper respiratory tract infections
- And/ or performance scale 2: symptomatic, normal activity

Stage 3

- Weight loss (>10%)
- Unexplained chronic diarrhoea lasting for longer than a month
- Unexplained prolonged fever (intermittent or consistent) > 1 month
- Oral candidiasis
- Vulvovaginal candidiasis
- Vulvovaginal candidiasis, chronic (>1 month) or poorly responsive to therapy
- Oral hairy leukoplakia
- Pulmonary tuberculosis within the last year
- Severe bacterial infections such as pneumonia
- And/or performance scale: bedridden < 50% of the day in the last month

Stage 4 (AIDS)

- HIV wasting syndrome
- Pneumocystis carinii Pneumonia (PCP)
- Toxoplasmosis of the brain
- Cryptosporidiosis with diarrhoea > 1 month
- Cryptococcosis (extrapulmonary)
- Cytomegalovirus (CMV)
- Herpes simplex virus
- Progressive multifocal leukocephalopathy
- Oesophageal, tracheal, bronchial or pulmonary
- Atypical mycobacteriosis (disseminated)
- Non-typhoid salmonella septicaemia
- Extrapulmonary tuberculosis
- Lyphoma (most commonly Non-Hodgkin's lymphoma)

- Endemic mycosis

Name: _____

Number: _____

DOB: _____ R/S: _____

Hospital: GSH or Somerset

Date of **HIV** diagnosis: _____ Transmission: _____

CD4 lymphocyte count : 1. _____ Date: 1. _____
2. _____ 2. _____
3. _____ 3. _____

Total lymphocyte count: 1. _____ 1. _____
2. _____ 2. _____
3. _____ 3. _____

Date of **TB** diagnosis: _____
Method of **TB** diagnosis:

	TEST	SPECIMEN
Culture		
Smear		
Histology		

Site of **TB**: 1. Pulmonary 2. Extrapulmonary(meningitis, nodal, pericardial)

CXR: Glands- Y/ N
Infiltrate- Y/ N; if Y- diffuse, cavities, other
Effusion- Y/ N

Date last seen: _____

Date of death: _____

Cause of death: _____

Name:

Other HIV-related morbidity (WHO staging): YES/ NO

Condition	Date- @ TB diagnosis or w/in 6 mo **prior**	Date- **after** TB diagnosis
1) Acute retroviral infection, PGL		
2) Weight loss <10% Minor Mucocutaneous (inc. zoster) Recurrent URI		
3) Weight loss >10% Unexplained chronic diarrhoea, >1 month Unexplained fever, >1 month Thrush/OHL Severe bacterial infections (pneumonia)		
4) HIV wasting syndrome PCP (Pneumocystis carinii pneumonia) Toxoplasmosis of the brain Cryptosporidiosis w/ diarrhoea, >1 month Cryptococcosis, extrapulmonary Cytomegalovirus (organ other than liver, spleen, nodes Herpes simplex virus, mucocut >1 month or visceral Candidiasis of oesophagus, trachea, bronchi, or lungs Atypical mycobacteriosis, disseminated Non-typhoid salmonella septicaemia Kaposi's sarcoma HIV encephalopathy Cervical cancer Progressive multifocal leuko-encephalopathy Any disseminated endemic mycosis (histo..) Lymphoma Pneumonia, recurrent		

APPENDIX IV **DATA SHEET**

PATIENT NUMBER			CURRENT VISIT NO.		
DATE OF BIRTH			DEPENDANTS	SPOUSE	1
AGE				1 CHILED	2
				2 CHILDREN	3
GENDER	MALE	1		3 OR MORE	4
	FEMALE	2		NON	5
SEXUAL PREFERENCE	HOMOSEXUAL	1	EDUCATION	PRIMARY	1
	HETEROSEXUAL	2		SECONDARY	2
	BISEXUAL	3		TERTIARY	3
	UNKNOWN	4		UNKNOWN	4
OTHER RISK FACTORS	NONE	1	MEDICAL AID	YES	1
	IVDA	2		NO	2
	BLOOD T/F	3			
	MULTI. PARTNERS	4	REFERRED BY	GP	1
	OTHER			DAY HOSP.	2
				G.S.H	3
POPULATION GROUP	WHITE	1		RED CROSS	4
	COLOURED	2		TB HOSP.	5
	AFRICAN	3		POLLSMOOR	6
	ASIAN	4			
			OTHER		
MARITAL STATUS	SINGLE	1			
	MARRIED	2			
OCCUPATION STATUS	EMPLOYED	1			
	UNEMPLOYED	2	HEP BAG	POSITIVE	1
	PENSIONER	3		NEGATIVE	2
	HOMEMAKER	4		UNKNOWN	3
	DISABILITY	5			
	STUDENT	6	VDRL	POSITIVE	1
	OTHER	7		NEGATIVE	2
CURRENT / PREVIOUS EMPLOYMENT	PROFESSIONA / MANAGERIAL	1	TPHA	POSITIVE	1
				NEGATIVE	2
	WHITE COLLAR / MIDDLE MANAGEMENT	2			
			PLACE OF RESIDENCE		
	SKILLED/ARTISAN	3	PLACE OF ORIGIN		
	SEMI-SKILLED	4	PLACE OF DIAGNOSIS		
	UNSKILLED	5	DATE OF DIAGNOSIS		
	NOT APPLICABLE	6	DATE OF 1ST VISIT		

HOSPITAL NUMBER																	
VISIT	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
MONTH																	
DAY																	
WHO STAGING																	
CLINICAL STAGE																	
PERFORMANCE SCALE																	
WEIGHT																	
LABORATORY																	
CD4																	
TOTAL LYMPHOCYTE																	
HB																	
PLTS																	
ESR																	
MINOR HIV RELATED CONDITIONS																	
ORAL CANDIDIASIS (1)																	
PERSISTENT G. L. (2)																	
OHL (3)																	
DIARRHOEA UNEXPLAINED (4)																	
HERPES ZOSTER (5)																	
PULMONARY TB (6)																	
MAJOR HIV RELATED CONDITIONS																	
EXRA PULMONAR TB (1)																	
P.C.P. (2)																	
TOXOPLASMOSIS. CNS (3)																	
CRYPTOCOCCSI (4)																	
KAPOSI' S SARCOMA (5)																	
DRUGS																	
ZIDOVUDINE (1)																	
BACTRIM (2)																	
PENTAMIDINE (3)																	
TB DRUGS (4)																	
OUTCOME																	
DOCTOR'S INITIALS																	

The scheme used for stratifying the patents' sociodemographic status

Variable:	**Score**
1. Occupation status : 1. Employed. 2. Unemployed	 1 0
2. Employment status: 1. Professional/ managerial / White collar/ Meddle management/Skilled/ Artisan. 2. Semi-skilled/ Unskilled.	 1 0
3. Education level: 1. Secondary/tertiary 2. Primary/Uneducated.	 1 0
4. Medical aid coverage: 1. Covered. 2. Uncovered.	 1 0
5. Place of residence: 1. High socioeconomic area. 2. Lower socioeconomic area.	 1 0
6. Number of dependants: : 3. Two dependants or less. 4. Three dependents or more.	 1 0
Total (i.e. highest score)	**6**

The place of residence was categorized as a lower or higher "sociodemographic area" according to the classification system used by the Cape Metropolitan Council (CMC).

Based on the variables in the above table, higher socidemographic status was defined as a score equal to or higher than 4. This cut-off is similar to the "Best-off" criteria used by the CMC, except for the medical aid variable included in the above scheme.