OCCURRENCE AND DETERMINANTS OF TREATMENT FAILURE IN ANTIRETROVIRAL THERAPY AT TSHWANE DISTRICT HOSPITAL

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A research report submitted to the Faculty of Health Sciences, University of the Witwatersrand, Johannesburg, in partial fulfilment of the requirements for the degree of Master of Science in Pharmaceutical Affairs

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DECLARATION

I, Temitope Sokoya declare that this research report is my own work. It is being submitted for the degree of Master of Science in Pharmaceutical Affairs in the University of the Witwatersrand, Johannesburg. It has not been submitted before for any degree or examination at this or any other university.

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_____ day of _____, 2012

DEDICATION

This research is dedicated to the Almighty God who is the source of life and all things. To my dearest husband, Daremi, and my beloved sons, Toluniyin and Ibukunoluwa.

ABSTRACT

Objective: To determine the proportion of HIV+ patients who fail treatment on a yearly basis in a 5-year treatment cohort in Tshwane District Hospital and to determine the correlation of treatment failure with variables routinely measured at the clinic namely WHO stage, CD4 count, HIV viral load, age, gender, presence of concomitant diseases, concomitant medication and distance travelled to clinic.

Design: A retrospective study with an analytical component was conducted using the hospital records of adult patients receiving antiretroviral therapy in 2004 and followed for 5 years (until 2009) at the Tshwane District Hospital.

Methods: All adult patients receiving antiretroviral therapy in 2004 were identified and followed for the next 5 years till 2009 at Tshwane District Hospital. The proportion of patients that failed treatment yearly was calculated. Univariate analysis was used to compare all patients who failed at any time point with the patients who did not fail at all for all variables. A repeated measures logistic regression model was developed to determine the variables that impacted on the binary outcome, namely treatment failure or not.

Results: Of the 1104 adult patients who were attending the TDH Immunology clinic in 2004, 870 adults were receiving ARVs. 333 patients (38.28 %) experienced treatment failure throughout the study period. 6.9 % (60/870) of the study population failed virologically. 307 of the 870 patients (35.29 %) failed treatment immunologically. 102 patients (11.72 %) experience treatment failure at the 12 month time point, 37 patients(4.49 %) at the 24 month time point, 57 patients(6.93 %) at 36 month time point, 101 patients(12.27 %) at the 48 month time point and 140 patients (7.01 %) failed treatment at 60 month time point. Univariate analysis showed significant correlation between treatment failure and non-adherer, interrupting treatment, defaulted treatment, viral load at baseline, 12, 24, 36, 48, 60 months, and CD4 count at baseline, 12, 24, 36, 48, 60 months. In the multivariate analysis, there was a significant association between short term stoppage of treatment (STSTOP) (coefficient ratio = 1.41; $p<0.001$), long term stoppage of treatment (LTSTOP) (coefficient ratio = 3.24; $p<0.001$), transfers from other health institutions (coefficient ratio = 1.96; $p<0.001$), regimen (coefficient ratio = -0.1734) and treatment failure. The change in log viral load at 12 months from baseline (LOGVLBL12) (coefficient ratio =-1.7145; $p<0.001$) was highly significant for reaching the end point - treatment

failure. Older patients were less likely to fail treatment (coefficient ratio = -0.0517, p<0.001) and patients with an advance stage of the disease (WHO stage 3 or 4) were at a lower risk of failing treatment (coefficient ratio = -0.4175; P=0.008). The CD4 count was significant in the univariate analysis P<0.01) and XTGEE (coefficient ratio =- 0.0001; p<0.001). There was no significant correlation between gender, place of residence, employment status and treatment failure.

Conclusion: More than one–third of the patients receiving treatment in TDH failed treatment within the 5 year study period. The determinants of treatment failure are age, WHO stage, transfer from other institutions, short term stoppage of treatment, long term stoppage of treatment, CD4 cell count and the level of viral suppression within the first year of treatment (LOGVLBL-12). This study reinforces the need for identifying high risk patients earlier in treatment in order to implement strategies that might strengthen adherence to treatment.

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ABBREVIATIONS

3TC	Lamivudine
AIDS	Acquired Immune Deficiency Syndrome
ART	Antiretroviral Treatment
ARV	Antiretroviral
AZT	Zidovudine
CD4	Cluster of Differentiation 4
D4T	Stavudine
ddl	Didanosine
DOH	Department of Health
EFV	Efavirenz
HAART	Highly Active Antiretroviral Therapy
HIV	Human Immunodeficiency Virus
LPV/r	Lopinavir/ritonavir
NNRTI	Nonnucleoside Reverse Transcriptase Inhibitors
N(t)RTI	Nucleo(t)side Reverse Transcriptase Inhibitors
NVP	Nevirapine
PI	Protease inhibitor
TB	Tuberculosis
TDF	Tenofovir
TDH	Tshwane District Hospital
WHO	World Health Organisation
XTGEE	Cross-Sectional Time-series Generalized Estimation Equations

CHAPTER 1: INTRODUCTION AND LITERATURE REVIEW

1.1 BACKGROUND

1.1.1 HIV – the global perspective

The year 2011 marks 30 years since the discovery of AIDS. The Joint United Nations Programme on HIV/AIDS (UNAIDS) estimated that the number of people living with HIV has slightly increased from 33.3 million in 2009 to just over 34 million in 2010[68]. While approximately 10 % of the world's population lives in sub-Saharan Africa, an enormous 64 % of all people living with HIV live in this region -including 77 % of all women living with HIV. Children are a particularly vulnerable group with high rates of mother-to-child- transmission as well as the impacts of ill-health and death of parents. The UNAIDS estimated that at the end of 2009, about 2.5million children were living with HIV globally with about 360000 in Nigeria and 330000 in South Africa[48].

The overall growth of the epidemic has stabilized in recent years. The sub-Saharan Africa- region carries the greatest burden of the epidemic with the highest adult prevalence between 5 % and 20 % in some countries. The prevalence in East Asia (adult prevalence <0.1 %) have remained relatively stable and are still largely concentrated among high-risk groups. Conversely, the number of people living with HIV in Eastern Europe and Central Asia has doubled since 2000 with adult prevalence at 0.8 %.

The new UNAIDS strategy 2011–2015 aims to advance global progress in achieving country set targets for universal access to HIV prevention, treatment, care and support and to halt and reverse the spread of HIV and contribute to the achievement of the Millennium Development goals by 2015. By the end of 2010, antiretroviral drugs that extend lives reached more people in resource-constrained countries—6.6 million—than ever before.

1.1.2 HIV in South Africa – the scope of the epidemic

In 2009, the UNAIDS estimated that about 5.6million people were living with HIV and AIDS in South Africa and that the adult prevalence was 17.8 %. Of the 5.6million people living with HIV/AIDS, about 3.3million are women and 330000 are children. Approximately 310000 people died of AIDS in South Africa in 2009[68]. South Africa has the largest antiretroviral therapy programme in the world, but given it also has the world's largest epidemic, access to treatment is low. At the end of 2009, an estimated 37 percent of infected people were receiving treatment for HIV according to the latest WHO guidelines (2010)[52]. The challenges of universal access to ARVs include cost and procurement problems, sustained availability of ARVs to prevent interruption of treatment due to stock-outs, inaccessible regular diagnostic tests, overburdened health workers due to lack of investment in the health services and the rapid scale-up of ART, recurrent transport costs which challenge continued access and adherence to ART, continuing stigma and discrimination of people in need of treatment, and lack of adherence and nutritional support for ART users.

The 2007-2011 National Strategic Plan aimed for one quarter of all people to take a test every year by 2011, with the proportion of those ever taking a test increasing to 70 percent[20]. In 2010 the South African Government launched a major HIV counselling and testing campaign (HCT) [41]. The aim of HCT campaign was to test a total of 15million people throughout South Africa by the end of June 2011 and has had a notable impact on the availability and uptake of HIV testing and treatment[41].

In mid-2011, following the launch of the HCT campaign in early 2010, the number of people on antiretroviral treatment had increased significantly from 923,000 in February 2010 to 1.4 million in May 2011[30].

1.2 INTRODUCTION

Use of effective antiretroviral therapy (ART) has led to major improvement in the health of HIV-infected populations[33]. The development of potent antiretroviral therapy (in the form of Highly Active Antiretroviral Therapy (HAART)) has substantially reduced AIDS-related morbidity and mortality[26]. For most people treated to date, ART has been based on a combination of drugs from two of the three original

classes; non-nucleoside reverse transcriptase inhibitors (NNRTIs); nucleo(t)side reverse transcriptase inhibitors [N(t)RTIs]; and protease inhibitors (PIs). The key indicators of the degree of success of the national ART programme include the proportion of patients on therapy that achieve HIV viral load suppression to lower than detectable levels and the proportion of patients that attain a CD4 count >350 cells/ mm[36].

Formerly, the natural history of HIV infection was invariably unidirectional, progressively leading to acquired immunodeficiency syndrome (AIDS) and death, and the efficacy of therapy was determined by its ability to delay this fast progression[34]. The treatment of HIV infection started with the use of one antiretroviral drug (mono therapy) and then dual therapy and it was only in 1996 that it was realised that a combination of three antiretroviral drugs (triple therapy) could achieve an undetectable viral load. Today, the clinical prognosis of HIV infection has radically changed because of the widespread use of HAART[34]. Partly because most studies link plasma HIV -1 RNA levels with risk of clinical progression, the positivist goal of antiretroviral therapy is now to reduce and maintain HIV – 1RNA levels below the lowest detectable level[55]. As the duration of infection increases, however, the mortality rate among HIV- infected patients increases compared with the general population[53]. This long-term excess mortality is likely to persist because antiretroviral therapy - related toxicity, non-adherence, and drug resistance, which may lead to treatment failure, are likely to increase with time on combination antiretroviral therapy.

Failure of therapy is usually defined in terms of lack of sufficient suppression of viral replication[34]. There are 3 kinds of treatment failure namely: virological failure, immunological failure and clinical failure. Increasing numbers of patients have experienced multiple episodes of virological failure, with those who initiated therapy with mono or dual nucleoside therapy before the HAART era, at particularly high risk[6]. Studies have shown that the lowest HIV-1 RNA levels achievable are required to obtain durable virological responses. Durability of virological and immunological responses should be understood as the major goal to improve the clinical prognosis of patients [34]. Evaluating the proportion of patients who have experienced treatment failure is important for understanding the likely durability of ART success.

1.3 LITERATURE REVIEW

1.3.1 Antiretroviral treatment rollout in South Africa

Antiretroviral treatment helps in controlling viral replication, maintaining immunologic function and long-term survival in HIV-positive individuals [40].

The South African Government has standardised the ARV regimens used in the pharmacological management of HIV and AIDS. This is stipulated in the National Antiretroviral Treatment Guidelines (DOH, 2004 and 2010) (Table 1).

Table 1: Recommended ARV regimens for adults before April 2010

Regimen	ARV Drugs
1a	Stavudine / lamivudine / efavirenz
1b	Stavudine / lamivudine / nevirapine
2	Zidovudine / didanosine / lopinavir/ritonavir

Source: South African National Antiretroviral Treatment Guidelines (DOH, 2004:6)

The criteria for ART initiation in adults and adolescents as stipulated in the national antiretroviral treatment guidelines 2004 was a CD4 count < 200cells/mm^3 irrespective of stage or WHO stage IV AIDS-defining illness, irrespective of CD4 count and patient willingness and readiness to take ART adherently.

South Africa HIV treatment guidelines were changed in March 2010. Table 2 shows the standardised national ART regimens for adults and adolescents from March 2010 (DOH, 2010).

Table 2: The standardised national ART regimens for adults and adolescents (DOH, 2010)

1st Line

All new patients needing treatment including pregnant women	TDF+3TC/FTC+ EFV/NVP	For TB co-infection EFV is preferred. For women of child bearing age, not on reliable contraception NVP is preferred.
Currently on d4T based regimen with no side-effects	D4T + 3TC + EFV	Remain on d4t if well tolerated. Early switch with any toxicity. Substitute TDF if at high risk of toxicity
Contraindication to TDF: renal disease	AZT + 3TC + EFV/NVP	

2nd line

Failing on a d4t or AZT-based 1st line regimen	TDF + 3TC/FTC + LPV/r	Virological failure must be followed by intensive adherence management, as resuppression is often possible. If repeat VL remains >1000 in 3months despite adherence intervention, switch.
Failing on a TDF-based 1st line regimen	AZT + 3TC + LPV/r	Virological failure must be followed by intensive adherence management, as resuppression is often possible. If repeat VL

		remains >1000 in 3months despite adherence intervention, switch.

Salvage

Failing any 2nd line regimen	Specialist referral	Virological failure on protease inhibitors is almost always due to non-adherence. Intensively exploring and addressing issues relating to causes of non-adherence will most often lead to resuppresion

TDF - Tenofovir, 3TC – Lamivudine, D4T – Stavudine, EFV – Efavurenz, NVP – Nevirapine, AZT – Zidovudine, LPV/r – Lopinavir/Ritonavir

1.3.2 What is Treatment Failure?

Failure of therapy is when the antiretroviral therapy fails to slow down the replication of the virus in the body; hence sufficient viral suppression is not achieved.

1.3.3 Types of treatment failure

a) **Virologic Failure** – is the inability to achieve maximal suppression of HIV replication (undetectable viral load) or the achievement of a maximal suppression followed by virological rebound [50]**.** According to the South African Antiretroviral Treatment Guidelines (2010), virological failure is define as a viral load >1000 copies/ml on two occasions, despite intensive adherence counselling. A study by Paredes et al found that the median time to virological failure was 19 months[34]. Virological failure is usually the earliest form of treatment failure experienced by patients failing treatment and is the most sensitive indicator of treatment failure. Patients with virologic failure usually progress to immunologic failure if they do not switch to a more

effective treatment regimen. Immunologic failure may be followed by clinical failure**.**

b) **Immunologic Failure** – can be variously defined as: the failure of the CD4 count to rebound, a return to below the pre-therapy baseline value or a fall in CD4 count to less than 50 % of the maximum CD4 count while on therapy or a persistent CD4 level below 100 cells/mm^3 [12]. This usually occurs as a consequence of virological failure but can also occur even if a very low or undetectable viral load is achieved [7]. The latter scenario is called a discordant treatment response and may be due to numerous reasons, for example, old age, failure of the thymus to produce more CD4 cells or underlying malignancy.

c) **Clinical Failure** – is characterized by AIDS-defining events. This describes the situation in which an individual exhibits disease progression in terms of new, recurrent, or progressing AIDS-related opportunistic infections or HIV-related symptoms such as weight loss, fatigue, and sweats, in a patient on ART [12]. Clinical failure has been used to monitor antiretroviral treatment efficacy in settings where CD4 count or viral load testing is unavailable, but is not a good indicator of treatment failure as it follows so long after virological failure.

1.3.4 Factors that may cause treatment failure

- **Poor adherence**: Adherence means taking medication on time, and taking the prescribed dose in the correct way. The close connection between adherence and viral load, CD4 counts, and mortality has been unequivocally demonstrated. Adherence levels of $\geq$ 95 % are required to maintain virologic suppression[56]. Common reasons given for poor adherence include adverse effects, excessive pill burden and/or dosing frequency, dietary restriction, or simply not having medication available or forgetting to take doses.
- **Drug resistance**: This means that an antiretroviral drug or combination of drugs cannot prevent or reduce HIV replication. The ability of HIV to mutate and reproduce itself in the presence of antiretroviral drugs is called HIV drug

resistance. Changes (mutation) in the virus might cause resistance. When HIV enters the body, it makes both perfect copies of itself called wild-type virus and copies with random mutations. Mutations that cause resistance occur naturally and randomly. Primary resistance occurs when a patient becomes infected with a resistant virus and is subsequently treated with a HAART regimen to which the virus is not susceptible[50]**.** Secondary resistance means resistance of HIV to antiretroviral drugs seen in individuals already receiving treatment. This is mostly due to insufficient drug levels – as when patients are non-adherent, suboptimal dosing, malabsorption, or drug-interactions – that allow viral replication in the presence of drug.

- **Pharmacokinetics:** This is the way a drug is absorbed, distributed, metabolized, and eliminated from the body. Variation in antiretroviral drug metabolism and poor drug absorption can lead to treatment failure.
- **Drug interactions:** Many over-the-counter and prescription medications, illegal drugs, herbs, vitamins and supplements interact with a lot of the antiretrovirals. Some antiretroviral also interact with each other. These interactions can lower the drug level of the antiretrovirals or cause them to become ineffective [2,5,24] .
- **Co- Morbidity:** This is the presence of medical conditions other than HIV infection. Concurrent use of medications for this condition and antiretrovirals may lead to additive side effects or drug interactions.
- **Inappropriate choice of antiretroviral agent:** Taking suboptimal treatments that were available before the current regimen such as monotherapy (single dose of Nevirapine) and dual-therapy of antiretroviral drugs.
- **Inadequate or inconsistent drug supply:** This can lead to suboptimal treatment levels in the body and consequently to the development of drug resistance and treatment failure.

Treatment failure should be suspected if progression of HIV disease continues following initiation of HAART. Early treatment failure is associated with a low CD4 count, low body mass index, and anaemia, but these markers are nonspecific and could reflect advanced HIV infection, co-infection, and /or malnutrition.

1.3.5 Diagnosing treatment failure

The viral load test measures the amount of HI virus in the blood. The HIV-1 viral load measurement indicates the number of copies of HIV-1 RNA per millilitre of plasma. HIV ultimately resides within cells, and the plasma measurement is only a proxy measure of the burden of infection and the magnitude of viral replication. It is used to assess the risk of disease progression and can help guide initiation of therapy. It is critical in monitoring virologic response to ART.

CD4 count is used to crudely assess the immune system of patients. When HIV infects humans, CD4 lymphocyte cells (also called T-cells or T-helper cells) are the primary targets of HIV. The virus becomes part of the cells, and when they multiply to fight an infection, they also make more copies of HIV. The CD4 count and the CD4 percentage mark the degree of immunocompromise. When someone is infected with HIV for a long time, the number of CD4 cells declines. This is a sign that the immune system is weakening. It is used to determine the risk of opportunistic illnesses, assess prognosis, and guide decisions about when to start antiretroviral therapy (ART).

The World Health Organisation (WHO) staging system for HIV disease is based on clinical symptoms, which may be used to guide medical decision making. It is an approach for resource limited settings and is widely used in Africa and Asia and has been a useful tool in monitoring progression to symptomatic HIV disease[69]. Following infection with HIV, the rate of clinical disease progression varies enormously between individuals. Many factors such as host susceptibility and immune function, health care and co-infections as well as factors relating to the viral strain, may affect the rate of clinical disease progression.

1.3.6 Switching of ART regimen

Symptoms of drug toxicity and of immune recovery are common immediately after initiation of ART, but usually resolve spontaneously. However, if severe or not resolving symptoms are due to drug side-effects, drug substitutions can be made easily and safely. Substitution of an offending drug may be done to solve drug toxicity. The offending drug can be replaced with another drug from the same class that does not have the same adverse effect (e.g. d4t to AZT). Treatment switches –

substituting all three drugs - are only made when there is virological failure (VL>1000 copies/ml on two occasions, despite intensive adherence counselling). Virological failure is almost always due to poor adherence, often due to poor attention by the clinicians to drug toxicity, or where social factors have not been addressed. It is not recommended to change treatment regimen when immunological failure occurs in the absence of virological failure. Patients who fail clinically (new opportunistic infections while on treatment) or immunologically (CD4 count dropping) without virological failure are unlikely to benefit from treatment switches, and require clinical assessment and appropriate investigation. Patients who have experienced virological failure with good adherence may be changed to second line therapy.

1.3.7 The rate of change of CD4 count and viral load

The viral load is expected to reduce gradually to undetectable levels after initiation of ART and this indicates suppression of viral replication. Conversely, the CD4 count is expected to increase gradually after initiation of ART and this signifies immune recovery. The slopes are the rates of change of CD4 count and HIV viral load over time. For the rate of change of CD4 count, a positive slope means there is an increase in CD4 count indicating immune recovery as expected while a negative slope signifies a decrease in the CD4 count. For the rate of change of viral load, a negative slope signifies a reduction in viral load which indicates viral suppression while a positive slope means there is an increase in the viral load indicating that viral suppression was not achieved. The rate of change of CD4 count/viral load was calculated by subtracting the CD4 count/viral load of the previous year from the CD4 count/viral load at a particular time point and then dividing the result by 12 months.

The rate of change of CD4 count and viral load in an individual have significant prognostic value in determining the time of progression to treatment failure and the durability of treatment. Surrogate markers can be used as predictors for the outcome of HIV infection and may provide guidance for initiation or change in antiretroviral therapy[57]. Both CD4 cell count and plasma HIV RNA level are markers of disease progression[62]. The rate of change of the viral load may be clinically useful as an important predictor of outcome such as predicting sustained virological response to treatment. Serial viral load measurements may prove to be a useful surrogate marker that might provide therapeutic guidance during all stages of the disease,

including clinically asymptomatic period. The cumulative viral load over time is a function of both the initial viral load and the rate of change of viral load and may thus be a sensitive measure of the progression of disease [23].

A public health approach to using combination ART is often used, which is designed to have the maximum clinical benefit on a population level by using combination ART, without necessarily providing individualized optimized treatment. A lack of resource and infrastructure may lead to difficulty in obtaining serial measurement of viral load and patient might remain on a virologically failing regimen as a consequence. For such patients in those setting where CD4 cell count are used to monitor patient, maintaining a CD4 level to reduce the risk of clinical disease progression despite virological failure become of utmost importance[61].

In patients with advanced stages of disease, and in treated patients, the rate of change of CD4 cell count may provide better information than viral load for long-term outcomes such as the incidence of new AIDS-defining illnesses or survival. The viral load changes may still be more useful in determining the efficacy of antiretroviral treatment and guiding therapeutic changes[58-60].

1.4 JUSTIFICATION

The advent of HAART has reduced the morbidity and mortality of HIV and improved the quality of life of people living with the virus. Since the launch of the HCT campaign by the South African government, more people have come to know their status and started on antiretroviral therapy. This is in line with the UNAIDS 2011-2015 strategy which aims to increase universal access to HIV prevention, treatment and care. However, core to the success of this programme is the durability of antiretroviral therapy. Inability to sustain treatment may lead to treatment failure and hence defeat the purpose. For antiretroviral therapy to work, patients must adhere to a daily regimen of ARVs for life. Interrupting treatment can result in the development of resistant strains of HIV making first line therapy no longer effective. In developing countries and resource limited settings, the classes of antiretroviral drugs available are limited. Therefore, keeping patients on treatment programmes is imperative. The numbers of patients on ART have increased rapidly in South Africa, but the programme has experienced deteriorating patient retention over time, particularly

due to apparent loss to follow-up and the increase in patient failing to follow up their ART is particularly worrying[10].

A continuous and sustained suppression of viral replication is required for prolonged clinical benefit [44]. Suboptimal viral suppression often leads to drug resistance and subsequently treatment failure and spread of resistance strains. Occurrence of treatment failure often has socio-economic implications because of the increased direct and indirect cost associated with starting more costly second-line treatment for patients. From the medical supplies depot in Gauteng province, the cost supply of antiretrovirals in 2011 regimen ranges from R78.92 to R131.04 for first-line treatment regimen depending on the drug combination and the cost of the second-line regimen is R341.87 which is almost 3 times that of the first-line regimen. The cost of a second-line treatment program remains extremely high and depends on factors such as the cost of the ARVs and price of testing (reagents and other costs) for viral load and resistance. Keeping patients on a failing regimen leads to the reversal of clinical improvements of patients to the pre-treatment state [29].

Knowledge of the factors that are predictive of treatment failure will aid in identifying patients that are at a higher risk of failure. Furthermore, in the absence of predictive factors, the scarce resources available may be wasted unnecessarily by using it for patients that are less likely to develop treatment failure [22]. This study provides valuable information about the clinical progression of patients on ART. Durability of virological response is one of the goals to improve the clinical prognosis of patients; hence monitoring the proportion of patients who have experienced drug failure is important for understanding the likely durability of antiretroviral therapy success. Understanding the proportion of patient failing treatment will also help in clinical decision making such as strengthening treatment adherence by counsellors and support groups.

This study utilized variables that are routinely measured in the clinic such as the HIV viral load and CD4 count in order to determine treatment failure. Other variables that are routinely collected such as the distance travelled to the clinic, WHO stage, age, gender and presence of concomitant disease, were assessed to find out if there is a relationship between treatment failure and these variables.

1.5 OBJECTIVES OF THE STUDY

The primary objectives of this study were:

a) To determine the proportion of HIV+ patients who fail treatment on a yearly basis in a 5-year treatment cohort in Tshwane District Hospital.

b) To determine the correlation of treatment failure with variables routinely measured at the clinic namely WHO stage, CD4 count, HIV viral load, age, gender, presence of concomitant diseases, concomitant medication and distance travelled to clinic.

The secondary objectives of the study were:

a) To determine the rate of change of CD4 counts over time

b) To determine the rate of change of HIV viral load over time

CHAPTER 2: METHODOLOGY

2.1 Study design

A retrospective study with an analytical component was conducted using the hospital records of adult patients receiving antiretroviral therapy in 2004 and followed for 5years (until 2009) at the Tshwane District Hospital. The demographic profile of the patients receiving antiretroviral treatment during this period was described and the study evaluated the occurrence and determinants of treatment failure in the patients that were receiving antiretroviral therapy at the hospital.

2.2 Study population and sampling

All adult patients who were receiving antiretroviral therapy in 2004 were eligible for inclusion in the study. There was an existing database that was developed for patients receiving treatment at the Immunology clinic of the Tshwane District Hospital. This database stretched over two years and was as accurate as the files would allow, thus, a pilot study and sampling was not required. All records for patients enrolled on HAART during this period were included.

2.3 Method

Since this was a record review, all the necessary patient information was obtained from the patients' medical records. Using the existing computerised database, which stretched over two years, the patients receiving HAART were separated from those who were not yet started on antiretroviral treatment as this formed the inclusion criterion of the study. These patients were allocated a specific identification number and all data of interest were reviewed and captured on the data extraction sheet (Appendix 1). The specific patient's clinical medical record was used to collect information on patient demographics, antiretroviral treatment history, laboratory findings and AIDS-defining events for the subsequent three years and then included in the data extraction sheet.

The following information was collected for each HIV positive patient receiving antiretroviral therapy during the study period.

- Age and Gender

- Place of residence - to be able to calculate the distance travelled to the clinic.
- Employment status (whether employed or not)
- CD4 count at baseline, 12month, 24month, 36month, 48month, 60month.
- Viral load at baseline, 12month, 24month, 36month, 48month, 60month.
- ARV treatment history (treatment regimen started on and treatment regimen changed to if a change was effected)
- HIV Staging – WHO defined stage at baseline, 12month, 24month, 36month, 48month and 60month.
- Concomitant diseases (Presenting disease) at baseline.
- Adherence factors (Non adherent patients, patients whose treatment was interrupted, patients that stopped treatment, defaulted or were transferred in from other centres.)

Based on the information gathered above the enrolled patients were subdivided into patients experiencing virological failure and immunological failures at each time point (12month, 24month, 36month, 48month, 60month)

2.4 Definition of variables

The outcomes of treatment were assessed using the different types of treatment failure definitions:

1.**Virological failure** - treatment failure defined by HIV viral load criteria - classified definite if the viral load is greater than 1000 copies/ml on two occasions at least 8 weeks apart, despite intensive adherence counselling, as this is in keeping with the most recent South African Antiretroviral Treatment Guidelines. The lowest level of detection of viral load was taken at 50 copies/ml (1.7log).

2. **Immunological failure** - treatment failure based on CD4 cell count criteria - defined according to WHO guidelines as either a return to below the pre-therapy baseline value or a fall in CD4 count to less than 50 % of the maximum CD4 count while on therapy or persistent CD4 level below 100 cells/ mm^3.

3. **Clinical failure** -treatment failure based on clinical events - defined as the occurrence of either a new or recurrent WHO defined stage 4 (WHO, 2010). Clinical failure was not used in this study since the WHO stage was not routinely captured at

each time point and since this was a retrospective review, could not be determined later.

The adherence factor was assessed based on the following definitions

- **Non adherer** –patients who missed more than 3 doses in a month according to self-report.
- **Interrupted** –patients whose ARVs were temporarily stopped (usually due to toxicity) and who were subsequently restarted on ARV treatment.
- **Stopped** –patients who permanently discontinued ARVs.
- **Defaulter** –patients who did not come back to the clinic for 3 months or longer.

2.5 DATA ANALYSIS

2.5.1 Statistical methods

Data were captured on an Excel spread sheet. Descriptive statistics were calculated, namely frequencies, means, medians, standard deviations, kurtosis, skewness, frequencies and proportions. Where groups were compared, the groups of interest were transferred to Statistix data analytical software version 9 (Analytical Software, Tallahassee, FL, USA). Two proportion hypothesis tests were used for binary data and chi square tests to determine the association between categorical variables. For continuous independent samples two-sample t-tests were used and Wilcoxon rank sum tests for nonparametric data. A p value of ≤ 0.05 was considered statistically significant. At each time point the patients who failed treatment were compared with the patients who did not fail treatment, for all variables, to find out if there was any correlation between them. All patients who failed at any time point were also compared with the patients who did not fail at all for all variables. A repeated measures logistic regression model was developed to determine the variables that impacted on the binary outcome, namely treatment failure or not.

2.5.2 Descriptive statistics for the study population

All HIV+ patients receiving ARVs at the hospital from 2004 until 2009 formed the study population. They were subdivided into treatment failure categories for analysis: virological failure at 12month, 24month, 36month, 48month, 60month, and

immunological failure at 12month, 24month, 36month, 48month, and 60month. Frequencies were calculated for the proportions of the study population that failed treatment at these time points. The same was done for gender, place of residence, employment status, WHO staging, non-adherers, defaulters, patient who interrupted treatment, patients who stopped treatment and patients who were transferred in.

2.5.3 Determination of proportion of HIV+ patients who fail treatment annually

The patients were divided into those experiencing virological failure and immunological failure at the different time points based on the viral load and CD4 counts, which were obtained from the hospital records. The HIV+ patients receiving ARVs who failed virologically were separated from those who failed immunologically at all time points. Proportions were calculated by dividing the number of HIV+ patients on ARVs who failed by the total number of patients receiving antiretroviral treatment in the hospital. Thereafter, the number of patients that experienced treatment failure, irrespective of virological or immunological, was divided by the total of patients receiving antiretroviral treatment. Further, all the patients who experienced any form of failure at any time point was divided by the total number of patients receiving antiretroviral treatment.

2.5.4 Variables routinely measured

Means, standard deviations, frequencies and proportions were calculated for gender, age, place of residence, employment status, and WHO stage for the whole population and for the patients that failed at each time point. Frequencies and proportions were also calculated for non-adherent patients, those who stopped treatment, defaulted in treatment or were transferred in from other health institutions. The groups that failed were then compared with those that did not using the appropriate hypothesis tests.

2.5.5 Determination of rate of change of CD4 count and viral load

The rate of change of CD4 count was calculated by subtracting the CD4 count of the previous year from the CD4 count at a particular time point and then dividing the result by 12 months, to get the rate of change per year.

The viral load values were transformed to logarithmic scale and the rate of change of viral load was calculated by subtracting the viral load of the previous year from the viral load at a particular time point and then dividing the result by 12 as before. Mean and standard deviations were calculated for the rates at each time point.

2.5.6 Logistic regression analysis

A causal logistic regression model was developed for predicting the likelihood of occurrence of treatment failure in this patient population. A dual strategy was used in developing this model, including a clinical hypothesis-driven approach in the form of causal directed acyclic graphs (DAGs)[14,70] and an inclusive variable selection statistical-criteria driven approach. Thus, both causal clinical reasoning and statistical considerations were employed in defining the model.

Initially, all variables considered in this study, including gender, age, employment status, place of residence, WHO stage, CD4 count viral load, presenting disease, regimen, non-adherence, interrupting treatment, defaulting treatment, stopping treatment and transfers were compared using univariate hypothesis tests between patients that did and did not fail. The variables that differed with p-values $\leq$ 0.2 were identified as potential candidates. This formed the first 'filter' in identifying an appropriate model.

A series of DAG iterations[14,70] representative of the evolving causal hypothesis were drawn using yEd graph editor version 3.8 (http://www.yWorks.com).

In terms of the repeated measures variables, covariance matrices were calculated to examine the nature of their correlation patterns across time. This was done for both CD4 count and log viral load using Stata version 11 (StataCorp, 4905 Lakeway drive, College station, TX 77845, USA). From these calculations it became apparent that CD4 count followed an autoregressive correlation structure over time, i.e. the correlation of observations with one another decreased as the time between them increased. This was reasonable in view of the steady rate of increase of CD4 counts over time, representing gradual immunological improvement following the initiation of ART. No similar pattern was readily observable for the log viral loads since these values normally dropped abruptly in the first year of ART.

Histograms were drawn and Shapiro-Wilk tests calculated to examine the normality of time-point data for CD4 and log viral loads. Spaghetti plots were used to compare CD4 count and log viral load for failure versus non-failure groups. Histograms were also drawn comparing the non-repeated measure variables for the failure versus non-failure groups.

Repeated measures logistic regressions were then calculated using the xtlogit command in Stata. Since this was a causal model an inclusive variable selection strategy was used for the variables identified in the failure versus non-failure univariate comparisons and the DAG iterations. It is noteworthy that the xtlogit command in Stata uses log-likelihood, maximum likelihood estimation (MLE) techniques for convergence and does not allow the specification of the covariance matrix. On the other hand, the xtgee command uses the robust 'sandwich' estimator and allows the specification of an autoregressive matrix.

In this study, fixed effects (xtlogit), mixed effects (xtlogit) and random or population effects regressions (xtlogit and xtgee) were used. A large number of iterations of regression demonstrated that random effects models gave the most parsimonious results. This was evaluated by comparing the log-likelihoods, the Wald Chi^2 and the p-values of each ensuing model. Note, if the random effects assumption holds, namely that population rather than individual- based effects are predominant, the random effects model is statistically more efficient than the fixed effects model[14]. The random effects models gave better p–values than any of the fixed effects models.

Using the statistical reasoning and the hypothesis-driven DAG iterations, 3 new variables were created: 1. LogvlBL-12, which was the difference between baseline log viral load and 12month log viral load. This was in view of the observation that the log viral loads demonstrated the most change in all groups in the first year of therapy. 2. STSTOP (short term stop) which was a composite variable for all patients whose treatment was interrupted or who were non adherent.3. LTSTOP (long term stop) which was also a composite variable, namely for those patients who completely stopped treatment or who defaulted.

2.6 ETHICAL CONSIDERATIONS

The study was approved by the University of the Witwatersrand Human Research Committee and the Tshwane Metsweding Region Research Ethics Committee. Permission was obtained from the Chief Executive Officer of the hospital to perform a retrospective review on hospital records. To ensure confidentiality, the patients' identity remained anonymous as no personal identifiers were captured into the data extraction sheet. All patients were given unique study numbers and the documents with the patients' details were kept private and confidential.

CHAPTER 3: RESULTS

3.1 Description of the entire population

3.1.1 Social demographics

The existing database had 1104 patients who were attending the TDH Immunology clinic; however 234 patients were excluded from this study because they were not taking ARVs at the start of the study. A total of 870 adults who were receiving ARVs were followed over the 5year study period. The laboratory results from the existing data system and hospital records were used to obtain the relevant information. As shown in table 3, the mean age of patients receiving ARVs was 42 years. Of the 870 patients, 272(31.34 %) were male and 596 (68.66 %) were female. Most of the patients were unemployed and the majority lived in Mamelodi, a predominantly black township to the east of Pretoria (Table 3).

Table 3: Social demographics of patients

Parameter	**total**
Total patients enrolled	870
Male	272 (31.34 %)
Female	596 (68.66 %)
Mean Age (range in years)	42.2 (20 years – 69 years)
Residence in:	
Pretoria	209 (24.08 %)
Cullinan	20 (2.30 %)
Eersterust	66 (7.60 %)
Mamelodi	396 (45.62 %)
Other	177 (20.39 %)
Employed	248 (28.67 %)
Unemployed	617 (71.33 %)

The total numbers for each demographic (gender, residence and employment status) differ from the total number of patients enrolled in the study (870) due to missing values in the files.

Of interest was the prevalence of female and unemployed patients. The patients were predominantly black and of low socioeconomic status.

3.1.2. WHO staging

WHO HIV staging classification (Appendix 2) was determined by a review of clinical parameters documented in the hospital records as at the time of commencing ARVs. Of the 870 patients, 21 patients (2.43 %) were classified stage 1, 107 patients (12.4 %) were classified as stage 2, 479 patients (55.5 %) were classified as stage 3, 256 patients (29.66 %) were classified as stage 4. Almost 85.2 % of patients receiving ARVs had advanced HIV disease and were classified as WHO stage 3 or 4 (table 4)

Table 4: WHO staging for patients receiving ARVs at TDH

WHO STAGE	NUMBER (N= 863)	PERCENTAGE (%)
1	21	2.43
2	107	12.4
3	479	55.5
4	256	29.66

Most patients were classified as WHO stage 3 or 4.

3.1.3 Virological and Immunological indices

As shown in table 5, the mean CD4 count increased gradually with the highest value achieved at the 60 month time point. There was a decrease in the viral load from the 12 month to the 24 month time point. There was no difference in the mean viral load at 36 month, 48 month and 60 month (Table 5).

Table 5: Mean CD4 count and mean viral load of patients receiving ARVs at TDH

Time point	Mean CD4 count (cells/mm^3)	Mean Viral Load (copies/ml) and the log value in brackets
Baseline	109.57	281560.4 (5.039)
12month	289.75	5679.63 (1.881)
24month	407.59	548.4 (1.770)
36month	465.06	51.21 (1.699)
48month	516.52	56.61 (1.701)
60month	529.52	54.12 (1.702)

The mean CD4 count increased gradually over the 5 year period.

3.1.4 Adherence factors

Of the 870 adult patient receiving antiretroviral therapy at the hospital, 111 (13.59 %) patients missed more than 3 doses according to self report, 131 (16.93 %) patients stopped ARVs temporarily (usually due to toxicity) and subsequently restarted, 27 (3.34 %) stopped ARVs permanently, 120 (14.65 %) patients did not come back to the clinic for 3months or longer hence defaulted in treatment, and 129 (15.75 %) patients were transferred to the clinic from other health institutions.

Table 6: The adherence factors of patients receiving ARVs at TDH

Parameter	Number of patients	Percentage (%)
Non adherer	(N = 817)	
Yes	111	13.59
No	706	86.41
Interrupted	(N = 774)	
Yes	131	16.93
No	643	83.07
Stopped	(N = 808)	
Yes	27	3.34
No	781	96.66
Defaulted	(N = 819)	
Yes	120	14.65
No	699	85.35
Transferred	(N = 819)	
Yes	129	15.75
No	690	84.25

The total numbers for the parameters are not the same due to missing values in the files. The majority of patients did adhere to the therapy

3.1.5. Antiretroviral treatment

Most of the patients, almost 66 % were started on the d4t, 3tc, efv combination. Of the 536 patient who were started on the d4t, 3tc, efv combination, 86 patients were changed to azt, 3tc, efv combination during the 5year period.

Table 7: The treatment regimen of patients receiving ARVs at TDH

Treatment Regimen started on	Number	Percentage
d4t, 3tc, efv	536	65.93 %
d4t, 3tc, nvp	70	8.61 %
azt, 3tc, efv	37	4.55 %
azt, 3tc, nvp	15	1.85 %
azt, 3tc, ddi, kaletra	2	0.25 %
other.	67	8.24 %
d4t, 3tc., efv change to azt,3tc,efv	86	16.04%

3tc – Lamivudine, d4t – Stavudine, efv – Efavirenz, nvp – Nevirapine, azt – Zidovudine, ddi – Didanosine.

The majority of the patients were on d4t, 3tc,efv combination when they started ART.

3.2 Proportion of patient who failed treatment virologically at each time point

3.2.1. Virological failure at 12months

Of the 870 patients receiving antiretroviral therapy, 45 patients failed virologically at the 12 month time point. Most of these patients were female (60 %) and the mean age was 42 years. Fourteen patients (31.11 %) resided in Pretoria central region, 2 patients (4.44 %) in Cullinan, 2 patients (4.44 %) in Eersterust, 18 patients (40 %) in Mamelodi and 9 patients (20 %) in other areas. Almost 69 % of these patients were unemployed and about 89 % were classified as WHO stage 3 or 4. Of these 45 patients, 13 patients (28.89 %) missed more than 3 doses in a month according to self report, 9 patients (21.43 %) interrupted treatment temporarily, 1 patient (2.22 %) stopped ARVs permanently, 6 patients (13.33 %) did not come back to the clinic for 3 months or longer and 7 patients (15.56 %) were transferred in from other health institutions. 66.67percent of these patients were on the first line treatment regimen-d4t, 3tc,efv combination (see Table 8).

3.2.2. Virological failure at 24months

As shown in table 8, 14 patients experienced virological failure at the 24 month time point. Of these patients 69.23 % were female and the mean age was 40years. Most of these patients (57.14 %) lived in Mamelodi, 1 patient (7.14 %) lived in Pretoria central area, 2 patients (14.29 %) lived in Eersterust and 3 patients (21.43 %) lived in other areas. All the patients were unemployed and about 86 % were classified as WHO stage 3 or 4. Of these 14 patients, 5 patients (38.46 %) missed more than 3 doses in a month according to self report, 5 patients (35.71 %) interrupted treatment temporarily, 4 patients (28.57 %) did not come back to the clinic for 3 months or longer and 1 patient (7.14 %) was transferred in from another health institution. Nine of these patients (64.29 %) were receiving the first line treatment regimen- d4t, 3tc,efv combination (Table 8).

3.2.3 Virological failure at 48 months

Only 1 patient failed virologically at the 48month time point. This patient was a 38-year old female who resided in Eersterust. The patient presented with WHO stage 3 disease and was unemployed. The patient did not come back to the clinic for 3 months or more and hence defaulted on treatment. She was placed on a special treatment regimen when her ARTs were restarted.

3.2.4 Virological failure at 36months and 60months

None of the 870 patients failed virologically at the 36month and 60month time points.

Table 8: Description of virological failure population

Parameter	12month (N = 870)	24month (N = 824)	36month (N= 823)	48month (N = 823)	60month (N =823)
No of patients	45 (5.17 %)	14(1.70 %)	0	1(0.12 %)	0
Male	18(40 %)	4(30.77 %)	N/A*	0	N/A*
Female	27(60 %)	9(69.23 %)	N/A*	1(100 %)	N/A*
Mean Age (years)	41.69 (28-67)	40.29(27-57)	N/A*	38	N/A*
Residence in					
Pretoria	14(31.11 %)	1(7.14 %)	N/A*	0	N/A*
Cullinan	2(4.44 %)	0	N/A*	0	N/A*
Eersterust	2(4.44 %)	2(14.29 %)	N/A*	1(100 %)	N/A*
Mamelodi	18(40 %)	8(57.14 %)	N/A*	0	N/A*
Others	9(20 %)	3(21.43 %)	N/A*	0	N/A*
Employed	14(31.11 %)	0	N/A*	0	N/A*
Unemployed	31(68.89 %)	14(100 %)	N/A*	1(100 %)	N/A*
WHO stage					
1	1(2.22 %)	0	N/A*	0	N/A*
2	4(8.89 %)	2(14.29 %)	N/A*	0	N/A*
3	29(64.44 %)	9(64.29 %)	N/A*	1(100 %)	N/A*
4	11(24.44 %)	3(21.43 %)	N/A*	0	N/A*
Mean Viral Load(copies/ml)	108578.5	30128.57	N/A*	0	N/A*
Non adherer	13(28.89 %)	5(38.46 %)	N/A*	0	N/A*
Interrupted1	9(21.43 %)	5(35.71 %)	N/A*	0	N/A*
Stopped 1	1(2.22 %)	0	N/A*	0	N/A*
Defaulter	6(13.33 %)	4(28.57 %)	N/A*	1(100 %)	N/A*
Transferred	7(15.56 %)	1(7.14 %)	N/A*	0	N/A*
Regimen					
d4t, 3tc, efv	30(66.67 %)	9(64.29 %)	N/A*	0	N/A*
d4t, 3tc, nvp	4(8.89 %)	1(7.14 %)	N/A*	0	N/A*
azt, 3tc, efv	3(6.67 %)	1(7.14 %)	N/A*	0	N/A*
azt, 3tc, nvp	0	0	N/A*	0	N/A*
azt, ddi, kaletra	0	0	N/A*	0	N/A*
azt, 3tc, ddi, kaletra	0	0	N/A*	0	N/A*
other.	6(13.33 %)	3(21.43 %)	N/A*	1(100 %)	N/A*
d4t, 3tc, efv changed to azt,3tc,efv	2(4.44 %)	0	N/A*	0	N/A*

*** N/A – not applicable**

3.3 Proportion of patients who failed treatment immunologically at each time point

3.3.1 Immunological failure at 12 months

Of the 870 patients receiving antiretroviral therapy, 72 patients failed immunologically at the 12month time point. Forty two patients (58.33 %) were female while 30 patients (41.67 %) were male. The mean age was 39 years. Sixteen patients (22.54 %) resided in Pretoria central region, 3 patients (4.23 %) in Cullinan, 10 patients (14.08 %) in Eersterust, 28 patients (39.44 %) in Mamelodi and 14 patients (19.72 %) in other areas. 69.44percent of these patients were unemployed and about 86 % were classified as WHO stage 3 or 4. Of these 72 patients, 11 patients (17.74 %) missed more than 3 doses in a month according to self-report, 14 patients (24.14 %) interrupted treatment temporarily, 2 patients (3.28 %) stopped ARVs permanently, 14 patients (22.58 %) did not come back to the clinic for 3 months or longer and 9 patients (15 %) were transferred in from other health institutions. Thirty five patients (60.34 %) were on the first line treatment regimen- d4t, 3tc, efv combination (Table 9).

3.3.2 Immunological failure at 24months

As shown in table 9, 23 patients experienced immunological failure at the 24 month time point. Of these patients 60.9 % were female and the mean age was 40years. Most of these patients (69.57 %) lived in Mamelodi, 5 patients (21.74 %) lived in Pretoria central area, 1 patient (4.35 %) lived in Eersterust and 1 patient (4.35 %) lived in another area. Almost 87 % of the patients were unemployed and about 91 % were classified as WHO stage 3 or 4. Of these 23 patients, 8 patients (36.36 %) missed more than 3 doses in a month according to self-report, 12 patients (57.14 %) interrupted treatment temporarily, 1 patient (4.55 %) stopped ARVs permanently, 5 patients (22.73 %) did not come back to the clinic for 3 months or longer and 5 patients (22.73 %) were transferred in from other health institutions. Fifteen of these patients (68.8 %) were receiving the first line treatment regimen - d4t, 3tc, efv combination (Table 9).

3.3.3 Immunological failure at 36months

At the 36month time point, 57 patients receiving ARVs failed immunologically. Most of these patients were female (73.68 %) and the mean age was 39 years. Eleven patients (20 %) resided in Pretoria central area, 1patient (1.82 %) in Cullinan, 9 patients (16.36 %) in Eersterust, 26 patients (47.27 %) in Mamelodi and 8 patients (14.55 %) in other areas. Almost 75 % of these patients were unemployed and 85.45 % were classified as WHO stage 3 or 4. Of these 57 patients, 6 patients (13.04 %) missed more than 3 doses in a month according to self-report, 11 patients (24.44 %) interrupted treatment temporarily, 1 patient (2.17 %) stopped ARVs permanently, 13 patients (28.26 %) did not come back to the clinic for 3 months or longer and 11 patients (23.40 %) were transferred in from other health institutions. 55.56percent of these patients were on the first line treatment regimen - d4t, 3tc, efv combination (Table 9).

3.3.4 Immunological failure at 48months

One hundred adult patients of the study population experienced immunological failure at the 48 month time point. Of these patients 73.74 % were female and the mean age was 40 years. 47 patients (47 %) lived in Mamelodi, 21 patients (21 %) lived in Pretoria central area, 3 patients (3 %) lived in Cullinan, 11 patients (11 %) lived in Eersterust and 18 patients(18 %) lived in other areas. 73.47 % of the patients were unemployed and 81.81 % were classified as WHO stage 3 or 4. Of these 100 patients, 22 patients (23.4 %) missed more than 3 doses in a month according to self-report, 16 patients (18.18 %) interrupted treatment temporarily, 8 patients (8.6 %) stopped treatment permanently, 30 patients (31.91 %) did not come back to the clinic for 3 months or longer and 17 patients (18.48 %) were transferred in from other health institutions. 62 of these patients (67.39 %) were receiving the first line treatment regimen - d4t, 3tc,efv combination (Table 9).

3.3.5 Immunological failure at 60months

As shown in table 9, 140 of the adult patients receiving ART failed immunologically at the 60 month time point. Most of these patients were female (68.57 %) and the mean age was 41 years. Thirty five patients (25.18 %) resided in Pretoria central area, 4 patients (2.88 %) in Cullinan, 12 patients (8.63 %) in Eersterust, 63 patients

(45.32 %) in Mamelodi and 25 patients (17.99 %) in other areas. Almost 73 % of these patients were unemployed and about 83 % were classified as WHO stage 3 or 4. Of these 140 patients, 16 patients (12.60 %) missed more than 3 doses in a month according to self-report, 18 patients (17.48 %) interrupted treatment temporarily, 4 patients (3.54 %) stopped ARVs permanently, 22 patients (17.19 %) did not come back to the clinic for 3 months or longer and 22 patients (17.32 %) were transferred in from other health institutions. 65.08 % of these patients were on the first line treatment regimen - d4t, 3tc,efv combination (Table 9).

Table 9: Description of immunological failure population at each time point

Parameter	12months (N=870)	24months (N=824)	36months (N=823)	48months (N=823)	60months (N=823)
No of patients	72(8.28 %)	23(2.79 %)	57(6.93 %)	100(12.15 %)	140(17.01 %)
Male	30(41.67 %)	9(39.13 %)	15(26.32 %)	26(26.26 %)	44(31.43 %)
Female	42(58.33 %)	14(60.90 %)	42(73.68 %)	73(73.74 %)	96(68.57 %)
Mean Age (range in years)	39.47(21-67)	40.52(26-55)	38.72(21-60)	40.15 (20-60)	41.22(21-67)
Residence in					
Pretoria	16(22.54 %)	5(21.74 %)	11(20 %)	21(21 %)	35(25.18 %)
Cullinan	3(4.23 %)	0	1(1.82 %)	3(3 %)	4(2.88 %)
Eersterust	10(14.08 %)	1(4.35 %)	9(16.36 %)	11(11 %)	12(8.63 %)
Mamelodi	28(39.44 %)	17(69.57 %)	26(47.27 %)	47(47 %)	63(45.32 %)
Others	14(19.72 %)	1(4.35 %)	8(14.55 %)	18(18 %)	25(17.99 %)
Employed	22(30.56 %)	3(13.04 %)	14(25.45 %)	26(26.53 %)	37(27.01 %)
Unemployed	50(69.44 %)	20(86.96 %)	41(74.55 %)	72(73.47 %)	100(72.9 %)
WHO stage					
1	2(2.78 %)	0	0	4(4.04 %)	5(3.60 %)
2	8(11.11 %)	2(9.09 %)	8(14.55 %)	14(14.14 %)	19.13.67 %)
3	40(55.56 %)	15(68.18 %)	31(56.36 %)	46(46.46 %)	79(56.83 %)
4	22(30.56 %)	5(22.72 %)	16(29.09 %)	35(35.35 %)	36(25.90 %)
Mean CD4 Count (cells/mm^3)(standard deviation)	126.81 (100.50)	91.13(40.46)	237.56(106.62)	251.22(104.25)	277.29(90.11)
Non adherer	11(17.74 %)	8(36.36 %)	6(13.04 %)	22(23.40 %)	16(12.60 %)
Interrupted 1	14(24.14 %)	12(57.14 %)	11(24.44 %)	16(18.18 %)	18(17.48 %)
Stopped 1	2(3.28 %)	1(4.55 %)	1(2.17 %)	8(8.60 %)	4(3.54 %)
Defaulter	14(22.58 %)	5(22.73 %)	13(28.26 %)	30(31.91 %)	22(17.19 %)
Transferred	9(15 %)	5(22.73 %)	11(23.40 %)	17(18.48 %)	22(17.32 %)
Regimen					
d4t, 3tc, efv	35(60.34 %)	15(68.18 %)	25(55.56 %	62(67.39 %)	82(65.08 %)
d4t, 3tc, nvp	4(6.90 %)	4(18.18 %)	7(15.56 %)	9(9.78 %)	16(12.70 %)
azt, 3tc, efv	6(10.34 %)	0	2(4.44 %)	2(2.17 %)	2(1.59 %)
azt, 3tc, nvp	0	0	3(6.67 %)	6(6.52 %)	3(2.38 %)
azt, ddi, kaletra	0	0	0	0	0
azt, 3tc, ddi, kaletra	0	0	0	0	0
other.	4(6.90 %)	1(4.55 %)	4(8.89 %)	9(9.78 %)	14(11.11 %)
d4t, 3tc,efv changed to azt,3tc,efv	9(15.52 %)	2(9.09 %)	4(8.89 %)	4(4.35 %)	9(7.14 %)

3tc – Lamivudine, d4t – Stavudine, efv – Efavirenz, nvp – Nevirapine, azt – Zidovudine, ddi – Didanosine.

3.4 Proportion of patients who failed treatment (both virological and/or immunological) on a yearly basis

3.4.1 Treatment failure at 12months

One hundred and two patients experience treatment failure at the 12month time point. Of these 102 patients, 45 patients (44.12 %) experienced virological failure, 72 patients (70.59 %) experienced immunological failure and 15 patients experienced both virological failure and immunological failure. The mean age was 40years. 59 patients (57.84 %) were female and most of these patients lived in Mamelodi. About 86 % of these patients had WHO stage classification of 3 or 4. Sixty nine patients (67.65 %) were unemployed. Of these 102 patients, 20 patients (21.74 %) missed more than 3 doses in a month according to self-report, 22.99 % interrupted treatment temporarily, 3.3 % stopped ARVs permanently and 17 patients (18.48 %) did not come back to the clinic for 3months or longer (Table 10). Forty five patients (97.83 %) died within the first year of treatment. Though there were some missing data, this shows that majority of the death occurrence occurred within the first year of treatment.

3.4.2 Treatment failure at 24months

As shown in table 10, 37 patients failed treatment at the 24month time point. 14 patients (37.84 %) failed virologically, while 23patients (62.16 %) failed immunologically. None of the patients had both virological and immunological failure. Most of these patients were female (64.86 %) and the mean age was 42 years. Almost 92 % of these patients were unemployed, 62.16 % lived in Mamelodi and about 89 % of them were classified as WHO stage 3 or 4. Of these 37 patients, 12 patients (35.29 %) missed more than 3 doses in a month according to self-report, 16 patients (48.48 %) interrupted treatment temporarily, 1 patient (2.94 %) stopped ARVs permanently and 8 patients (22.86 %) did not come back to the clinic for 3 months or longer.

3.4.3. Treatment failure at 36months

At 36 month time point, 57 adult patients receiving ARVs failed treatment. No patients failed virologically at this time point, only immunological failure was seen at this time point. Most of these patients were female and the majority of them lived in

Mamelodi. The mean age was 39 years, 74.55 % were unemployed and about 85 % were classified as WHO stage 3 or 4. Of these 57 patients, 6 patients (13.04 %) missed more than 3 doses in a month according to self-report, 11 patients (24.44 %) interrupted treatment temporarily, 1 patient (2.17 %) stopped ARVs permanently, 13 patients (28.26 %) did not come back to the clinic for 3months or longer and 11 patients (23.40 %) were transferred in from other health institutions (Table 10).

3.4.4 Treatment failure at 48months

Of the 823 adult patients receiving ARVs at TDH, 101 patients failed at the 48month time point. One hundred patients failed immunologically while only one patient failed virologically. The mean age was 40 years, 75 % were female and most of these patients resided in Mamelodi. Of these patients, 82 % were classified WHO stage 3 or 4 and 72 patients (72.73 %) were unemployed. Twenty two patients (23.40 %) missed more than 3 doses in a month according to self-report, 16 patients (18.60 %) interrupted treatment temporarily, 8 patients (8.79 %) stopped ARVs permanently and 30 patients (31.91 %) did not come back to the clinic for 3 months or longer (Table 10).

3.4.5. Treatment failure at 60months

As shown in table 10, one hundred and forty patients failed treatment at 60month time point. Only immunological failure was experienced by these patients. 68.57 % of these patients were female and the mean age was 41 years. About 83 % were classified as WHO stage 3 or 4 and almost 73 % of these patients were unemployed. Of these 140 patients, 16 patients (12.60 %) missed more than 3 doses in a month according to self-report, 18 patients (17.48 %) interrupted treatment temporarily, 4 patients (3.54 %) stopped ARVs permanently, 22 patients (17.19 %) did not come back to the clinic for 3 months or longer and 22 patients (17.32 %) were transferred in from other health institutions (Table 10).

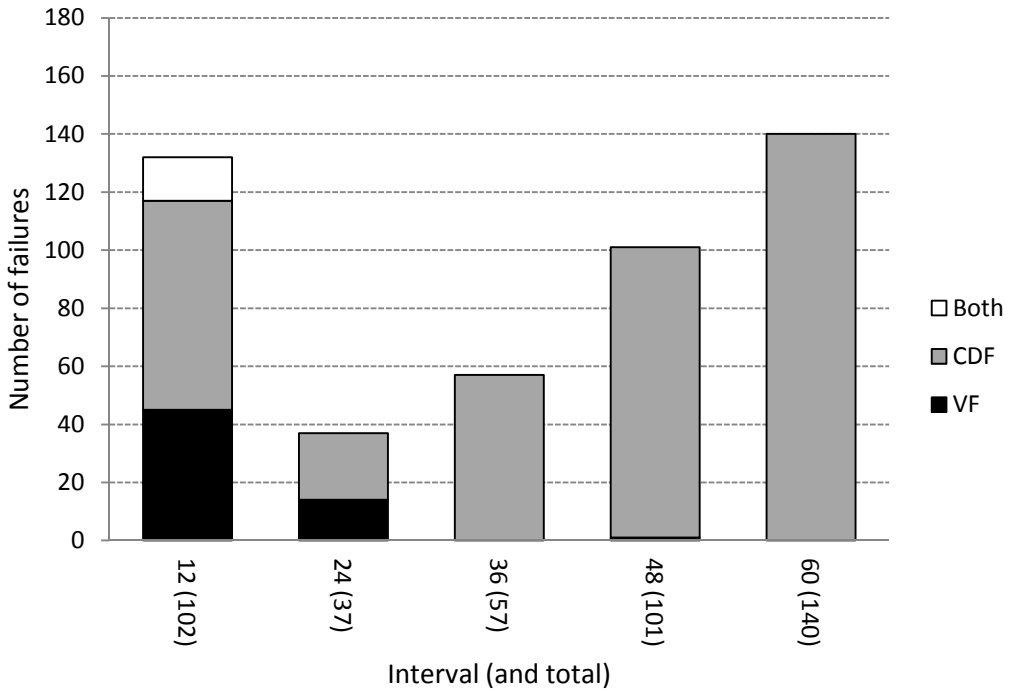
Table 10: Description of treatment failure population at each time point

Parameter	12month (N = 870)	24month (N = 824)	36month (N = 823)	48month (N = 823)	60month (N= 823)
No of patients	102(11.72 %)	37(4.49 %)	57(6.93 %)	101(12.27 %)	140(17.01 %)
Virological failure	45 (5.17 %)	14(1.61 %)	0	1(0.11 %)	0
Immunological failure	72(8.28 %)	23(2.64 %)	57(6.55 %)	100(11.49 %)	140(16.09 %)
Both failures	15(1.72 %)	0	0	0	0
Male	43(42.16 %)	13(35.14 %)	15(26.32 %)	25(25 %)	44(31.43 %)
Female	59(57.84 %)	24(64.86 %)	42(73.68 %)	75(75 %)	96(68.57 %)
Mean Age (range in years)	40.23(21-67)	42.16(26-57)	38.72(21-60)	40.37(20–60)	41.22 (21 -67)
Residence in					
Pretoria	27(26.73 %)	7(18.92 %)	11(20 %)	21(20.79 %)	35(25.18 %)
Cullinan	3(2.97 %)	0	1(1.82 %)	3(2.97 %)	4(2.88 %)
Eersterust	11(10.89 %)	3(8.11 %)	9(16.36 %)	12(11.88 %)	12(8.63 %)
Mamelodi	39(38.61 %)	23(62.16 %)	26(47.27 %)	47(46.53 %)	63(45.32 %)
Others	21(20.79 %)	4(10.81 %)	8(14.55 %)	18(17.82 %)	25(17.99 %)
Employed	33(32.35 %)	3(8.11 %)	14(25.45 %)	27(27.27 %)	37(27.01 %)
Unemployed	69(67.65 %)	34(91.89 %)	41(74.55 %)	72(72.73 %)	100(72.99 %)
WHO stage					
1	2(1.96 %)	1(2.78 %)	0	5(5 %)	5(3.60 %)
2	12(11.76 %)	3(8.33 %)	8(14.55 %)	14(14 %)	19.13.67 %)
3	60(58.82 %)	23(63.89 %)	31(56.36 %)	48(48 %)	79(56.83 %)
4	28(27.45 %)	9(25 %)	16(29.09 %)	33(33 %)	36(25.90 %)
Non adherer	20(21.74 %)	12(35.29 %)	6(13.04 %)	22(23.40 %)	16(12.60 %)
Interrupted 1	20(22.99 %)	16(48.48 %)	11(24.44 %)	16(18.60 %)	18(17.48 %)
Stopped 1	3(3.3 %)	1(2.94 %)	1(2.17 %)	8(8.79 %)	4(3.54 %)
Defaulter	17(18.48 %)	8(22.86 %)	13(28.26 %)	30(31.91 %)	22(17.19 %)

Table 11: Description of the types of treatment failure at each time point

	12month	24month	36month	48month	60month	Total
Number	102	37	57	101	140	437(100 %)
Virological failure	45	14	0	1	0	60 (13.73 %)
Immunological failure	72	23	57	100	140	392 (89.70 %)
Both failures	15	0	0	0	0	15 (3.43 %)

There were 437 instances of failure. 60 were virological failure, 392 were immunological failure and only 15 instances showed an overlap of both virological and immunological failure.



CDF – Immunological failure VF – Virological failure

Figure 1: Histogram showing the types of treatment failure at each time point

The above histogram shows that virological failure was only experienced in the 12month, 24month and 48month time points with a progressive decrease over time. However, for immunological failure, there was a sharp decrease in the proportion of patients that failed for the 12month time point to the 24month time point, but then gradually increased to the highest level at the 60month time point. As shown in table 11, there were a total of 437 instances of failure throughout the study period. Of these 437 instances, 60 were virological failure and 392 were immunological failure. Only 15 instances showed an overlap of both virological and immunological failure.

3.5 Total number of patients who failed treatment during the study period

Separating the patients who failed treatment at any time from those who did not fail treatment showed that; of the 870 adult patients receiving ARVs at TDH, 333 patients failed treatment at any time over the study period while 537 patients did not fail treatment at all. Though there were 437 instances of treatment failure (Table 11), only 333 patients failed treatment throughout the study period. This is due to the fact that some patients failed treatment more than once during the study period.

Table 12: Failure occurrence in the patients who failed treatment

Failure occurrence	1	2	3	4	Total
No of patients	248	72	11	2	333
Percentage (%)	74.47	21.62	3.3	0.6	100

Most patients failed treatment only once throughout the study period.

Table 12 shows the proportion of patients who experienced treatment failure once, twice or more. 74.47 % (248 patients) of the proportion of patients who failed treatment, failed once throughout the 5 year study period. 21.62 % (72 patients) of the failing population failed twice, 3.3 % (11 patients) failed 3 times and 0.6 % (2 patients) failed 4 times over the 5 year study period.

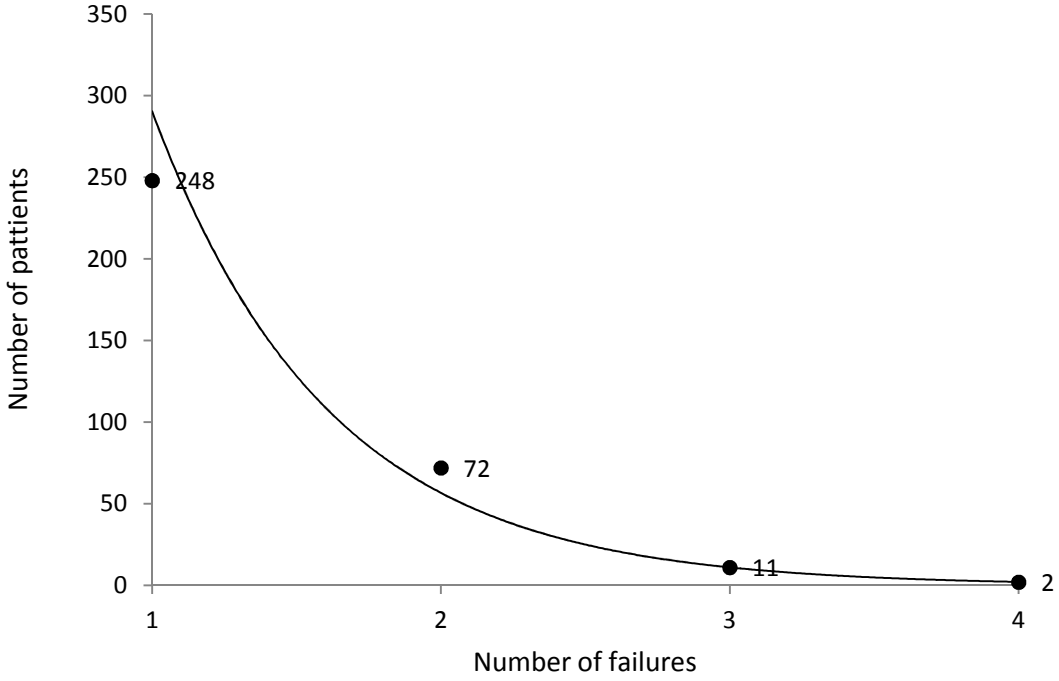


Figure 2: Graph showing failure occurrence in all patients

3.5.1 Social demographics

A total of 333 adults who were receiving ARVs over the 5 year study period experienced treatment failure at one or more time points. As shown below, the mean age of patients who failed treatment was 42 years. Of the 333 patients, 107 (32.33 %) were male and 224 (67.67 %) were female. Most of the patients were unemployed and the majority lived in Mamelodi, a township to the east of Pretoria.

Table 13: Social demographics of patients who failed treatment at TDH

Parameter	Number (percentage)
Total Patients	333
Male	107 (32.33 %)
Female	224 (67.67 %)
Mean Age(range in years)	41.54 (20 – 67)
Residence in	
Pretoria	77 (23.26 %)
Cullinan	10 (3.02 %)
Eersterust	30 (9.06 %)
Mamelodi	152 (45.92 %)
Others	62 (18.73 %)
Employed	86 (25.98 %)
Unemployed	245 (74.02 %)

3.5.2 WHO staging

Of the 333 patients, 9 patients (2.74 %) were classified as stage 1, 42 patients (12.77 %) were classified as stage 2, 187 patients (56.84 %) were classified as stage 3 and 91 patients (27.66 %) were classified as stage 4. 84.5 % of patients receiving ARVs had advanced HIV disease and were classified as WHO stage 3 or 4.

Table 14: WHO staging for patients that failed

WHO STAGE	NUMBER(N =329)	PERCENTAGE (%)
1	9	2.74
2	42	12.77
3	187	56.84
4	91	27.66

3.5.3 Virological and Immunological indices

As shown in table 15, the mean CD4 count increased gradually with the highest mean CD4 count attained at 48 months after which it slightly decreased at 60 months. There was a decrease in the viral load from baseline to the 24 month time point. The mean viral load at 36months, 48months and 60 months was relatively similar (Table 15).

Table 15: Mean CD4 count and mean viral load of patients who failed treatment

Time point	Mean CD4 count (cells/mm^3)	Standard deviation of mean CD4 count	Mean Viral Load (copies/ml) and the log value in bracket	Standard deviation of mean viral load.
Baseline	126.74	143.51	276849.1 (5.063)	331898.5
12month	282.17	182.65	14716.45 (2.111)	87730.83
24month	418.60	210.60	1335.97 (1.853)	13126.95
36month	452.98	196.07	52.91 (1.701)	35.30
48month	469.28	218.35	66.73 (1.707)	257.02
60month	441.26	201.52	55.24 (1.710)	48.18

3.5.4 Adherence Factors

Of the 333 adult patients who experienced treatment failure at the hospital, 56 (18.18 %) patients missed more than 3 doses according to self-report, 65 (22.41 %) patients stopped ARVs temporarily (usually due to toxicity) and subsequently restarted, 12(3.91 %) stopped ARVs permanently, 67 (21.61 %) patients did not come back to the clinic for 3 months or longer and hence defaulted on treatment, and 55 (17.74 %) patients were transferred to the clinic from other health institutions (Table 16).

Table 16: The adherence factors of patients who failed treatment at TDH

Parameter	Number of patients	Percentage (%)
Non adherer	(N = 308)	
Yes	56	18.18
Interrupted	(N = 290)	
Yes	65	22.41
Stopped	(N = 307)	
Yes	12	3.91
Defaulted	(N = 310)	
Yes	67	21.61
Transferred	(N = 310)	
Yes	55	17.74

The total numbers of patients for the parameters are not the same due to missing values in the files.

3.5.5 Antiretroviral treatment

Most of the patients, about 65 %, were started on d4t, 3tc, efv combination. Of the 200 patients who were started on d4t, 3tc, efv combination, 27 patients were changed to azt, 3tc, efv combination during the 5 year period (see table 17).

Table 17: Treatment regimen of patients that failed treatment at TDH

Treatment Regimen	Number (N = 307)	Percentage
d4t, 3tc, efv	200	65.15 %
d4t, 3tc, nvp	30	9.77 %
azt, 3tc, efv	17	5.54 %
azt, 3tc, nvp	7	2.28 %
azt, 3tc, ddi, kaletra	0	0 %
other.	26	8.47 %
d4t, 3tc., efv change to azt,3tc,efv	27	8.79 %

3tc – Lamivudine, d4t – Stavudine, efv – Efavirenz, nvp – Nevirapine, azt – Zidovudine, ddi – Didanosine.

3.6 Rate of change of CD4 count over time

3.6.1. Rate of change of CD4 count of the entire study population over time

The mean rate of change of CD4 cell count was highest at the 12-month time point with a 15.02 cell count change and then gradually decreased to the lowest value of 1.09 cell count change at the 60 month time point. Eight hundred and nineteen patients (94.14 %) had a positive slope at the 12-month time point; this number decreased gradually to 469 patients (53.91 %) at the 60-month time point. Subsequently, there was an increase over time in the number of patients with negative slopes annually with the maximum value reaching 401 patients (46.09 % at the 60 month time point (see table 18).

Table 18: The mean rate of change of CD4 count overtime

Time period	12-0 months	24-12 months	36-24 months	48-36 months	60-48 months
Mean rate of change of CD4 (cells/mm^3/month)	15.02	9.82	4.89	4.19	1.09
Number of positive slopes (N = 870)	819	712	598	523	469
Percentage (%)	94.14	81.84	68.74	60.11	53.91
Number of negative slopes (N = 870)	51	158	272	347	401
Percentage (%)	5.86	18.16	31.26	39.89	46.09

3.6.2. Rate of change of CD4 count of patients who failed treatment

There was a decrease in the mean rate of change of CD4 cell count overtime for both the patients who failed treatment and those who did not fail treatment. However, the mean rate of change of CD4 count for the patients who failed treatment was worse in that the mean rate of change of CD4 count actually became negative at the 60month time point. A significant difference between the patients who failed

treatment and those who did not fail treatment could however not be demonstrated. This was presumably in view of the large standard deviations, as shown in table 19.

Table 19.The mean rate of change of CD4 count of failed versus non-failure patients

Time period	12-0 month	24-12 month	36-24 month	48-36 month	60-48 month
Mean rate of CD4 change of patients who failed (cell/mm^3/month)	13	11	3	1	-2
Standard Deviation	15	16	19	25	27
Mean rate of CD4 change of patients who did not fail (cells/mm^3/month)	16	9	6	6	3
Standard Deviation	10	11	13	16	18

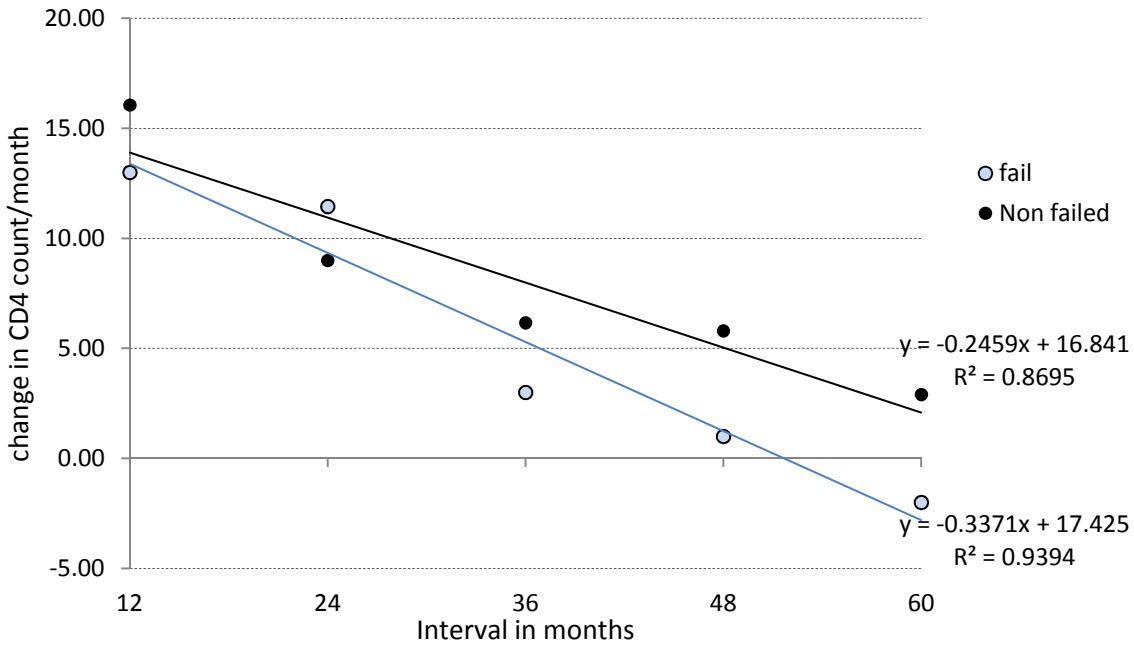


Figure 3: The mean rate of change in CD4 count /12 month period for failure versus non-failure patients

3.7 Rate of change of viral load over time

3.7.1 Rate of change of viral load of the entire study population over time

The mean rate of change in log viral load increased from a negative value at the 12 month time point to a positive value at the 48 month time point. However, the mean rate of change in log viral load remained the same at the 48 month and 60 month time points. Ninety eight percent (853 patients) of the 870 adult patients receiving ARVs had a negative slope at the 12 month time point. There was no significant difference in the percentage of patients with negative slopes at the other time points (see table 20).

Table 20: The mean rate of change of viral load over time

Time period	12-0 months	24-12 months	36-24 months	48-36 months	60-48 months
Mean rate of change of LOG VL. (copies/ml/month)	-0.263	-0.010	-0.006	0.000	0.000
Number of positive slopes (N = 870)	17	641	697	777	745
Percentage (%)	1.95	73.68	80.11	89.31	85.63
Number of negative slopes (N = 870)	853	229	173	93	125
Percentage (%)	98.05	26.32	19.89	10.69	14.37

3.7.2 Rate of change of log viral load of patients who failed treatment

The mean rate of change in log viral load for patients that failed treatment increased from a negative value at the 12 month time point to a positive value at the 48 month time point. This indicated that there was a slow rate of viral suppression over time in the patients that failed treatment. There was an increase in the mean rate of change of log viral load overtime for the patients who failed treatment. However, the mean rate of change of log viral load for patients that did not fail treatment remained the same over time, implying that the rate of viral suppression was constant over time.

Table 21: The mean rate of change of viral load of failure versus non-failure patients

Time period	12-0 month	24-12 month	36-24 month	48-36 month	60-48 month
Mean rate of LOG VL change of patients who failed (copies/ml/month)	-0.241	-0.027	-0.012	0.001	0.000
Standard Deviation	0.107	0.092	0.043	0.010	0.016
Mean rate of LOG VL change of patients who did not fail.(copies/ml/month)	0.000	0.000	0.000	0.000	0.000
Standard Deviation	0.069	0.022	0.019	0.009	0.013

There was a slow rate of viral suppression over time in the patients that failed treatment. The rate of viral suppression was constant over time for patients that did not fail treatment.

3.8 Hypothesis tests comparing failure versus non-failure patients for all variables

Table 22 presents the results of univariate comparisons between the failure and non-failure groups. There were significant differences between the two groups for the following variables: non-adherence, interrupting treatment, defaulted treatment, viral load at baseline, 12, 24, 36, 48, 60 months, and CD4 count at baseline, 12, 24, 36, 48, 60 months. Hence, there was a significant association ($p \leq 0.05$) between the clinical outcome, which is treatment failure, and the variables mentioned above. However, there was no significant association ($p > 0.05$) between gender of the patient, age of patient, place of residence, WHO stage, if a patient stopped treatment, if a patient was transferred from another health institution and whether a patient failed treatment or not.

Table 22: p-values for compared variables

Compared variables	p value≤0.05	p value >0.05
Gender all fail vs all not fail		0.6757
Age all fail vs all not fail		0.2543
Residence all fail vs all not fail		0.4666
Employment all fail vs all not fail		0.1938
WHO stage all fail vs all not fail		0.7655
Non adherer all fail vs all not fail	0.0040	
Interrupted all fail vs all not fail	0.0023	
Stopped all fail vs all not fail		0.6166
Defaulter all fail vs. all not fail	0.0008	
Transferred all fail vs. all not fail		0.2619
CD4 count baseline all fail vs. all not fail	<0.001	
CD4 count 12/12 all fail vs. all not fail	<0.001	
CD4 count 24/12 all fail vs. all not fail	<0.001	
CD4 count 36/12 all fail vs. all not fail	<0.001	
CD4 count 48/12 all fail vs. all not fail	<0.001	
CD4 count 60/12 all fail vs. all not fail	<0.001	
Viral load baseline all fail vs. all not fail	<0.001	
Viral load 12/12 all fail vs. all not fail	<0.001	
Viral load 24/12 all fail vs. all not fail	<0.001	
Viral load 36/12 all fail vs. all not fail	<0.001	
Viral load 48/12 all fail vs. all not fail	<0.001	
Viral load 60/12 all fail vs. all not fail	0.0021	

3.9 Results of logistic regression analysis

3.9.1 Analysis of covariance

Table 23: The p-value from analysis of covariance per time point of CD4 count and viral load

Parameter	P-value
CD4 count 0 versus Log VL 0,	**0.026**
CD4 count 12 versus Log VL 12	<0.01
CD4 count 24 versus Log VL 24	<0.01
CD4 count 36 versus Log VL 36	0.492
CD4 count 48 versus Log VL 48	**0.035**
CD4 count 60 versus Log VL 60	0.079

3.9.2 Covariance matrices

Table 24: The covariance matrix of CD4 count over the 5-year study period

	cd0	cd12	cd24	cd36	cd48	cd60
cd0	1.0000					
cd12	0.3682	1.0000				
	0.0000					
cd24	0.2842	0.5110	1.0000			
	0.0000	0.0000				
cd36	0.0399	0.2624	0.4202	1.0000		
	0.2397	0.0000	0.0000			
cd48	0.0597	0.1218	0.1412	0.1911	1.0000	
	0.0786	0.0003	0.0000	0.0000		
cd60	0.0368	0.0682	0.0182	0.0646	0.0749	1.0000
	0.2785	0.0444	0.5925	0.0569	0.0271	

The covariance matrix of CD4 count shows an autoregressive (AR1) correlation structure. The results that are nearer in time are more strongly correlated.

Table 25: The covariance matrix of the log viral load over the 5-years study period

	logvl0	logvl12	logvl24	logvl36	logvl48	logvl60
logvl0	1.0000					
logvl12	-0.0093	1.0000				
	0.7851					
logvl24	0.0213	0.1137	1.0000			
	0.5306	0.0008				
logvl36	-0.0325	-0.0410	0.0083	1.0000		
	0.3389	0.2265	0.8082			
logvl48	-0.0091	-0.0150	0.0292	0.5241	1.0000	
	0.7879	0.6590	0.3908	0.0000		
logvl60	0.0032	0.0984	-0.0067	-0.0006	-0.0755	1.0000
	0.9254	0.0037	0.8449	0.9855	0.0260	

The log viral load covariance matrix is unstructured hence there is no relationship across time.

3.9.3 Spaghetti plots for the CD4 count and viral load for the failed versus non failure groups

The spaghetti plot involves plotting a subject's values for the repeated outcome measure (vertical axis) versus time (horizontal axis) and connecting the dots chronologically. The spaghetti plot is useful for visually representing longitudinal data. The trajectories commonly overlap in a spaghetti plot, as both subjects and the magnitude of the outcome measure are displayed on the vertical axis[63].In the spaghetti plots below, the spikes in each trajectory represent the peak value of the CD4 count or viral load in each patient over time.

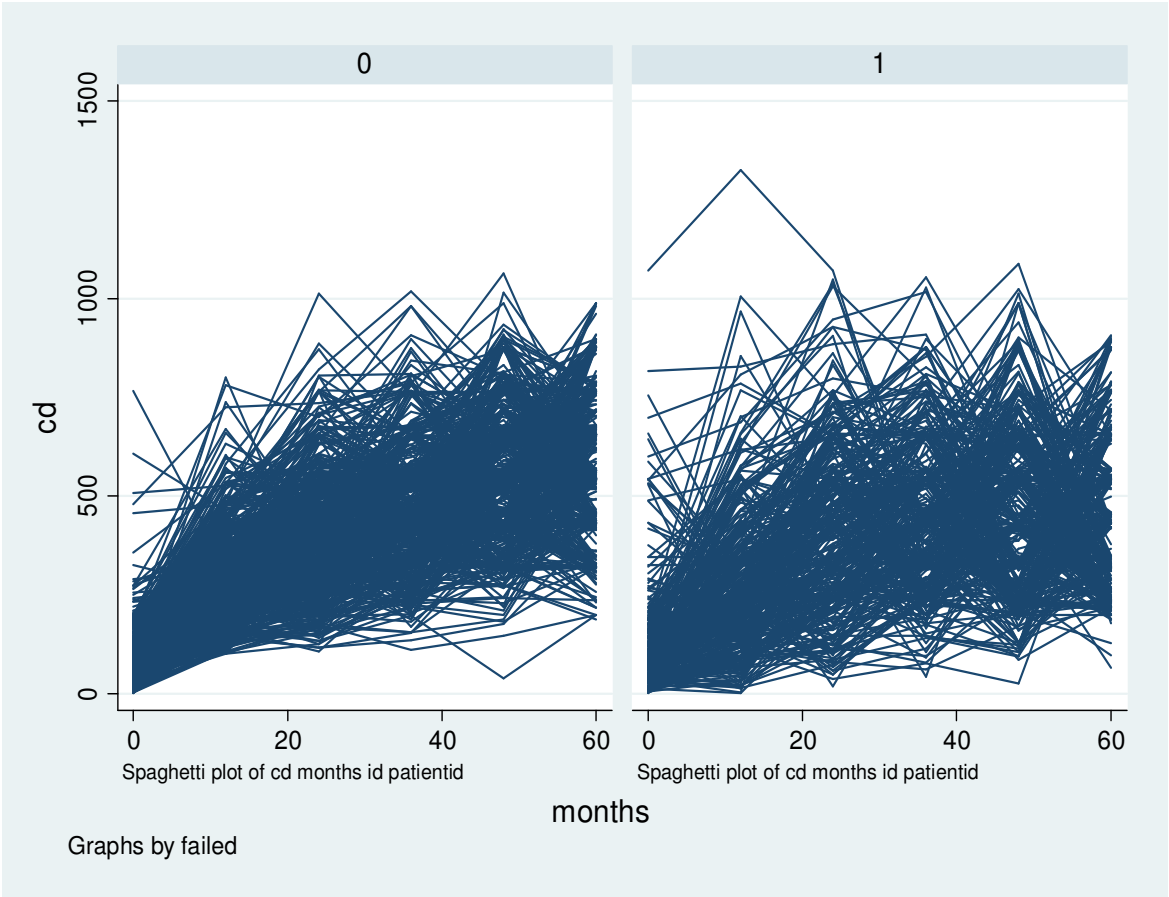


Figure 4: The spaghetti plot for CD4 count of non-failure patients (graph 0) and failure patients (graph 1)

In the spaghetti graph above the variation is greater in the failure group (graph 1) but it did not show any major difference in the CD4 count of the patients who failed treatment versus those that did not.

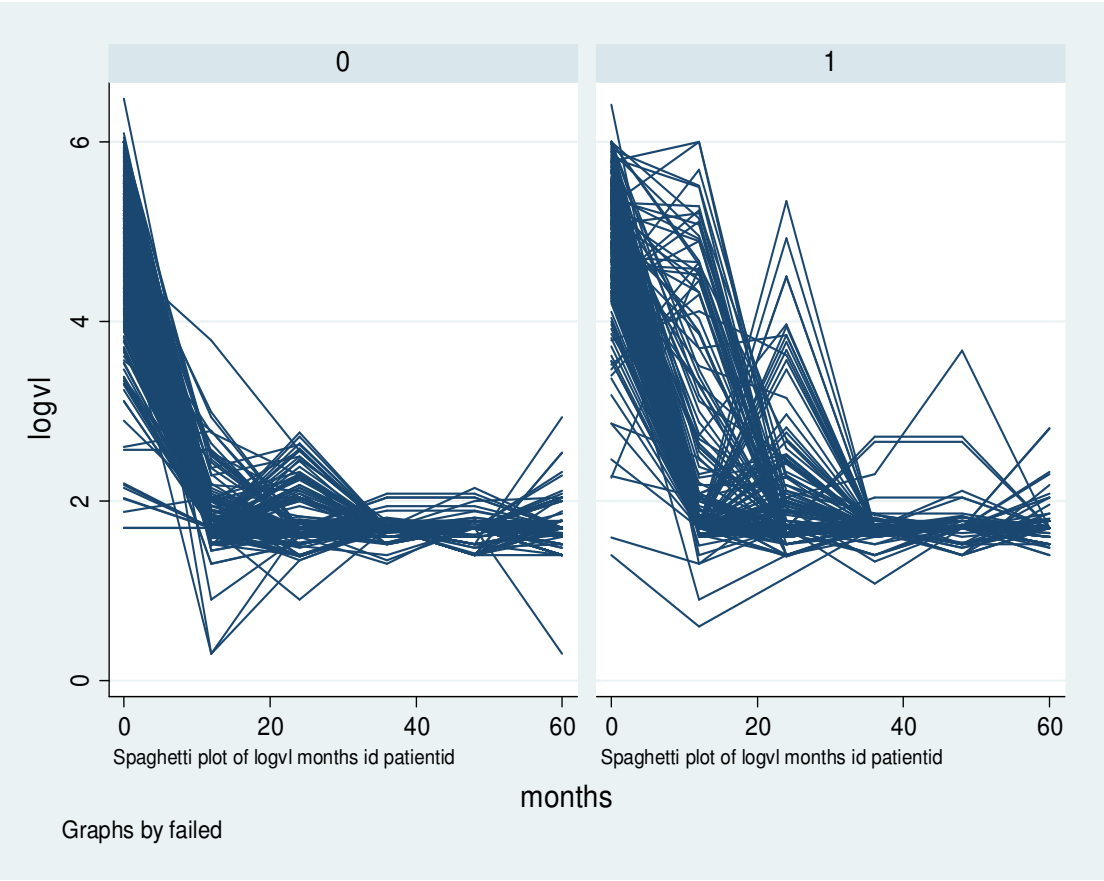


Figure 5: The spaghetti plot for log viral load of non-failure patients (graph 0) versus failure patients (graph 1)

Greater variation was observable in the graph of the failure group. A greater difference was detected at the 12 month time point. Consequently a new variable was created, namely LogvlBL-12, which is the difference between the baseline log viral load and 12 month log viral load. There was a significant association between treatment failure and LogvlBL12 ($p<0.01$). The histogram in figure 18 also shows a major difference between the groups of patients who failed treatment and those who did not fail treatment according to LogvlBL-12.

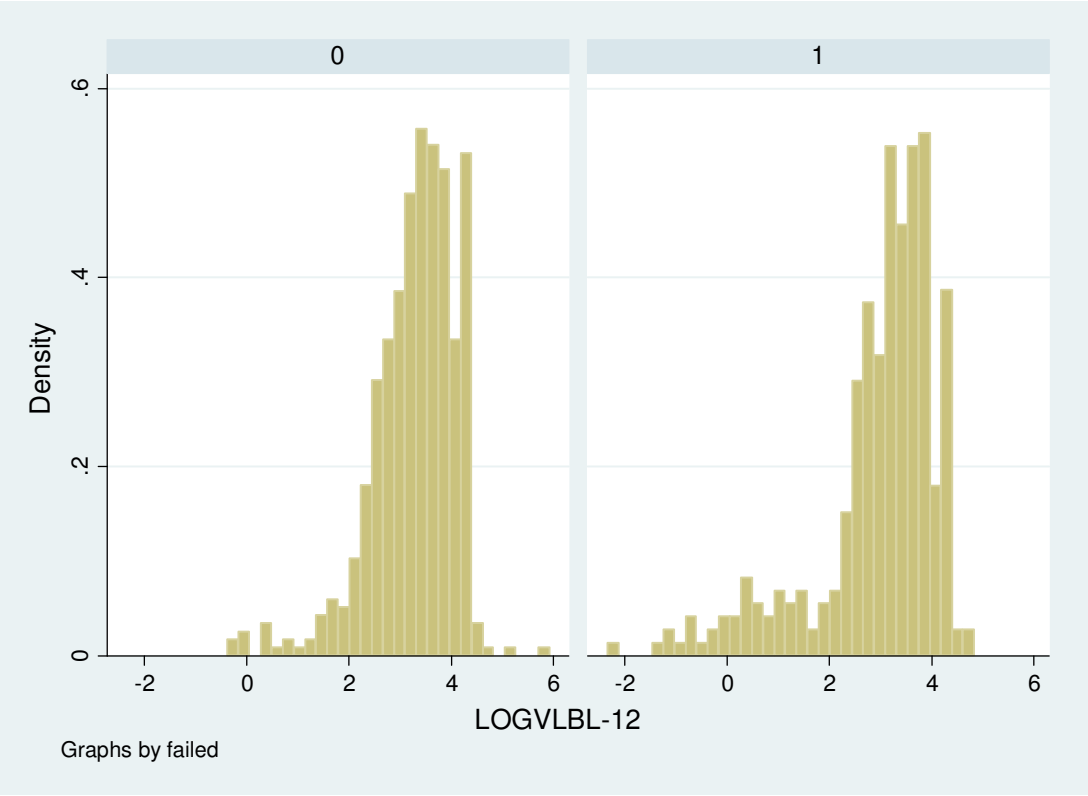


Figure 6: Histogram of LogvlBL-12 in the failure versus non failure groups

More patients with a negative value for LogvlBL-12 were present in the failure group. This indicated that there were more patients that did not achieve a reduction in their viral load within the first year of treatment, and eventually failed treatment. The rate of viral suppression within the first 12 months of treatment is lower in the patients who experienced treatment failure

3.9.4 Logistic regression

In this study, a causal model approach was used as opposed to a prognostic or prediction model since the objective was to find the underlying causes of treatment failure. The random effects model allows for between individual variability[27]. The basic idea in these models is that patient to patient variability is introduced by adding random effects as linear predictors in the regression relationship. Thus, in random effects models, heterogeneity and induced correlation can be thought of as arising from unobserved covariates[36]. The table below shows the results of the model of choice. The significance of the variables in predicting treatment failure is displayed according to p-values, odds ratios and beta coefficients.

Table 26: The result of xtlogit regression model of treatment failure

	ODDS RATIO (OR)	Beta coefficient	P-VALUE
Age	0.9495	-0.0517	**<0.001**
WHO	0.6587	-0.4175	**0.008**
Regimen	0.8408	-0.1734	**<0.001**
CD4 count	0.9995	-0.0005	0.273
LOGVLBL12	0.1800	-1.7145	**<0.001**
STSTOP	4.0882	1.4081	**<0.001**
LTSTOP	55165	3.2407	**<0.001**
Transfer	7.0969	1.9597	**<0.001**

STSTOP is a composite variable for non-adherer and/or interrupting treatment.

LTSTOP is a composite variable for defaulting and/or stopping treatment.

Age, WHO, regimen, the difference between the log viral load at 12month and baseline, long term stoppage of treatment, short term stoppage of treatment and transfers were significant predictors of treatment failure($p<0.05$) . The disadvantage of this model using the xtlogit command in Stata was the inability to specify the correlation matrix structure for the longitudinal measures, in this case CD4 count. The default was corr(exchangeable) – compound symmetry, and it was determined earlier that the CD4 count matrix was autoregressive 1.

Subsequently the xtgee command was used. The robust sandwich estimation algorithm resulted in faster convergence. This command also gives a population effects model but provides only beta coefficients not odds ratio.

Table 27: The result of the xtgee regression model showing p-value and coefficient ratio

	Beta coefficients	P-value
Age	-0.0109	0.195
Regimen	-0.0523	0.086
CD4 count	-0.0001	**<0.001**
LOGVLBL12	-0.3582	**<0.001**
STSTOP	0.3753	**0.039**
LTSTOP	0.9149	**<0.001**
Transfer	0.6386	**0.003**

Once the correlation structure of CD4 count (autoregressive 1) was specified the CD4 count panel variable became significant. However, both WHO stage and age lost significance. Thus, according to this model, the following variables were found to be significant predictors of treatment failure: CD4 count, the difference between the log viral load at 12month and baseline, long term stoppage of treatment, short term stoppage of treatment and transfers ($p \leq 0.05$). This seems to be a plausible model based on the clinical hypothesis.

3.9.5 Normality and distributional characteristics of the study population

Histograms and normal probability plots were used to examine the distributional characteristics of the population for the independent variables. If the sample conforms to a normal distribution, a plot of the rankits against the order statistics should result in a straight line. Systematic departure of the rankit plot from a linear trend indicates non-normality, as does a small value for the Shapiro-Wilk statistic.

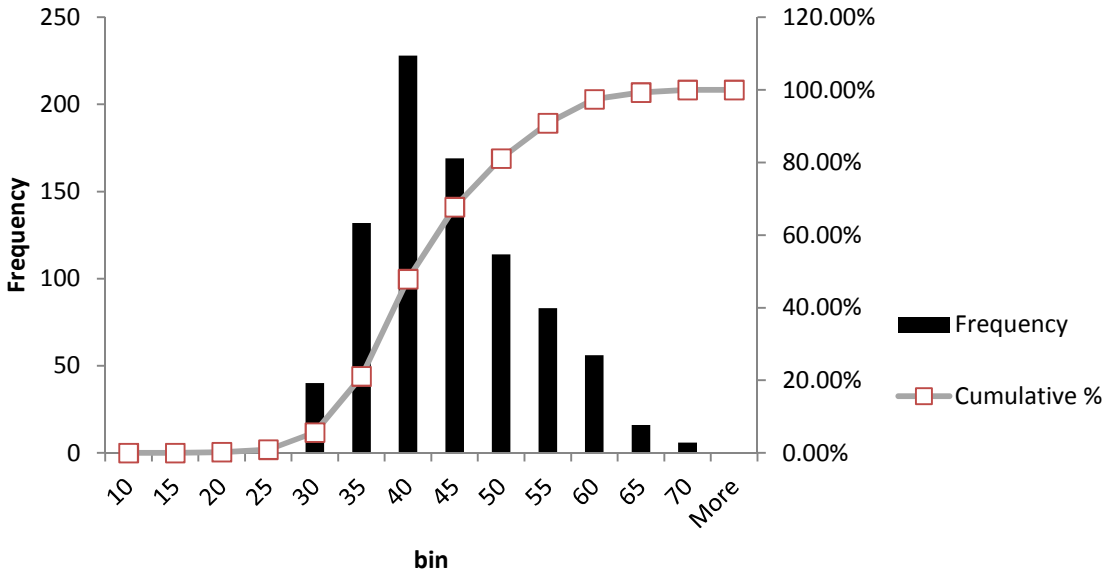


Figure 7: Histogram showing the distribution characteristics of age of the study population

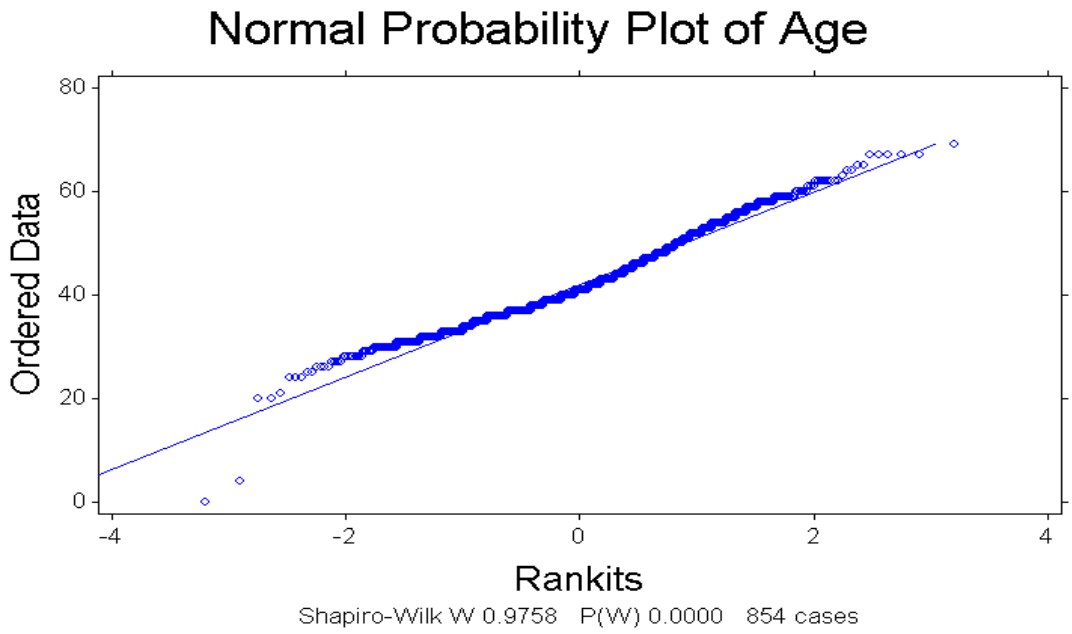


Figure 8: Normal probability plot of age

The histogram above shows that the study population had a greater proportion of older rather than younger people. Though the Shapiro-Wilk W value is close to one, we reject the null hypothesis that the data are normally distributed ($p<0.001$). The skewness of the data towards older patients would have impacted the regression model.

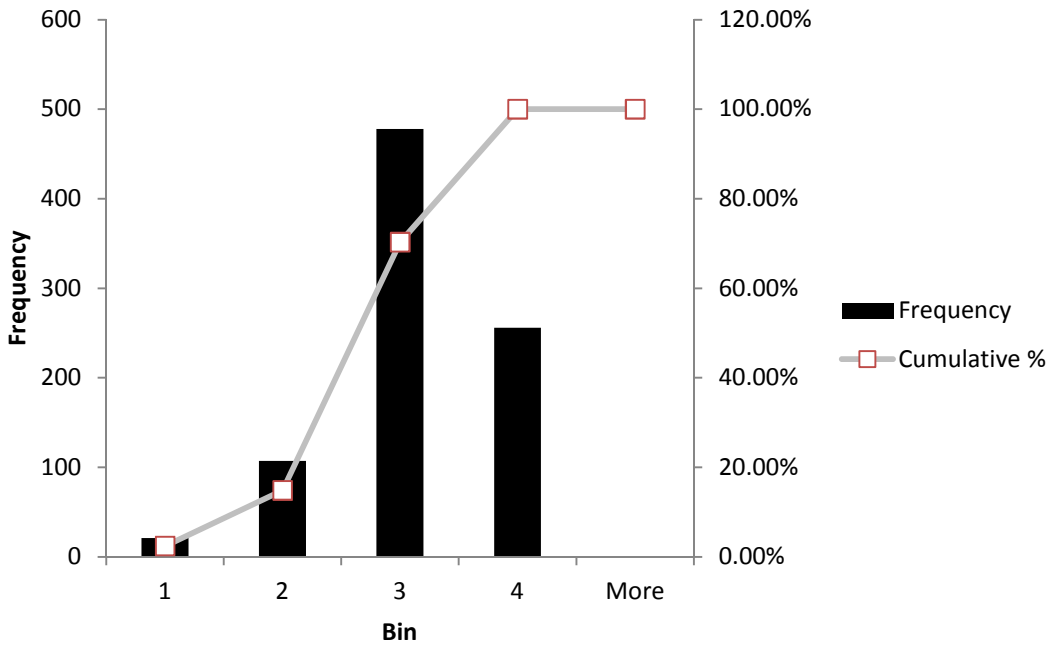


Figure 9.Histogram showing the distributional characteristics of WHO stage

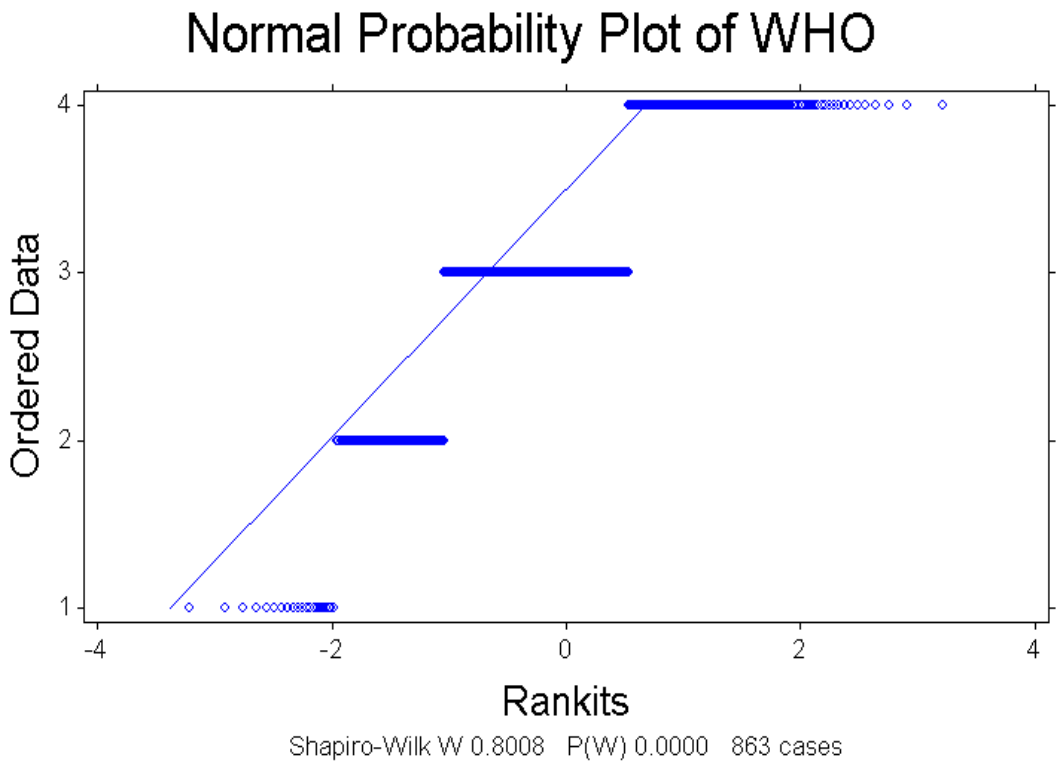


Figure 10: Normal probability plot of WHO stage

The result from the linear regression indicated that the patients that presented with a higher WHO stage at the time of initiation were less likely to fail treatment. The distribution of the sample population is not normal as shown in the histogram for WHO stage and the probability plot ($p<0.001$) and this may have had an impact on the regression analysis. In addition, it may also imply that more ill people were more likely to adhere to their treatment. Comparing the failed group versus not failed, there was a significant difference ($p=0.004$) in the adherence of the two groups which shows that adherence to treatment has a significant impact on the treatment outcome.

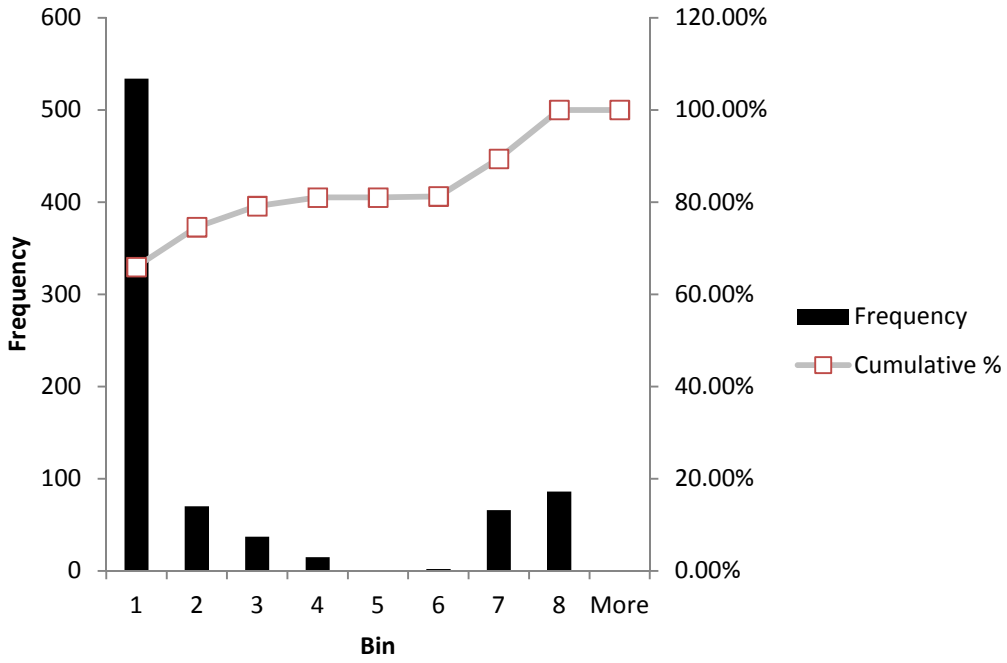


Figure 11: Histogram showing the distributional characteristics of treatment regimen

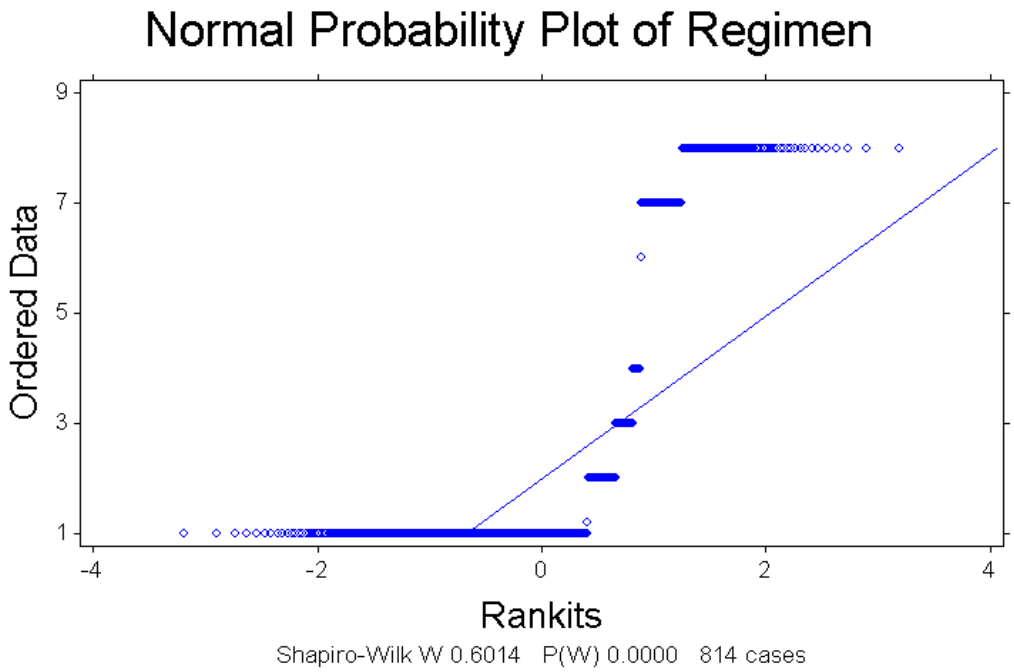


Figure 12: Normal probability plot of regimen

From the normal probability plot for regimen the Shapiro-Wilk W value is 0.6014 and $p<0.001$ hence we reject the null hypothesis that the data are normally distributed. The result of the regression model (beta coefficient = -01734) showed that the patient that were on second line regimen and those that were changed to another treatment regimen or special regimen were at a lower risk of experiencing treatment failure. This suggests that early detection of suboptimal suppression of virus by regular monitoring and appropriate clinical intervention is beneficial in delaying or preventing treatment failure. However, this study did not examine the effectiveness of the different treatment regimens.

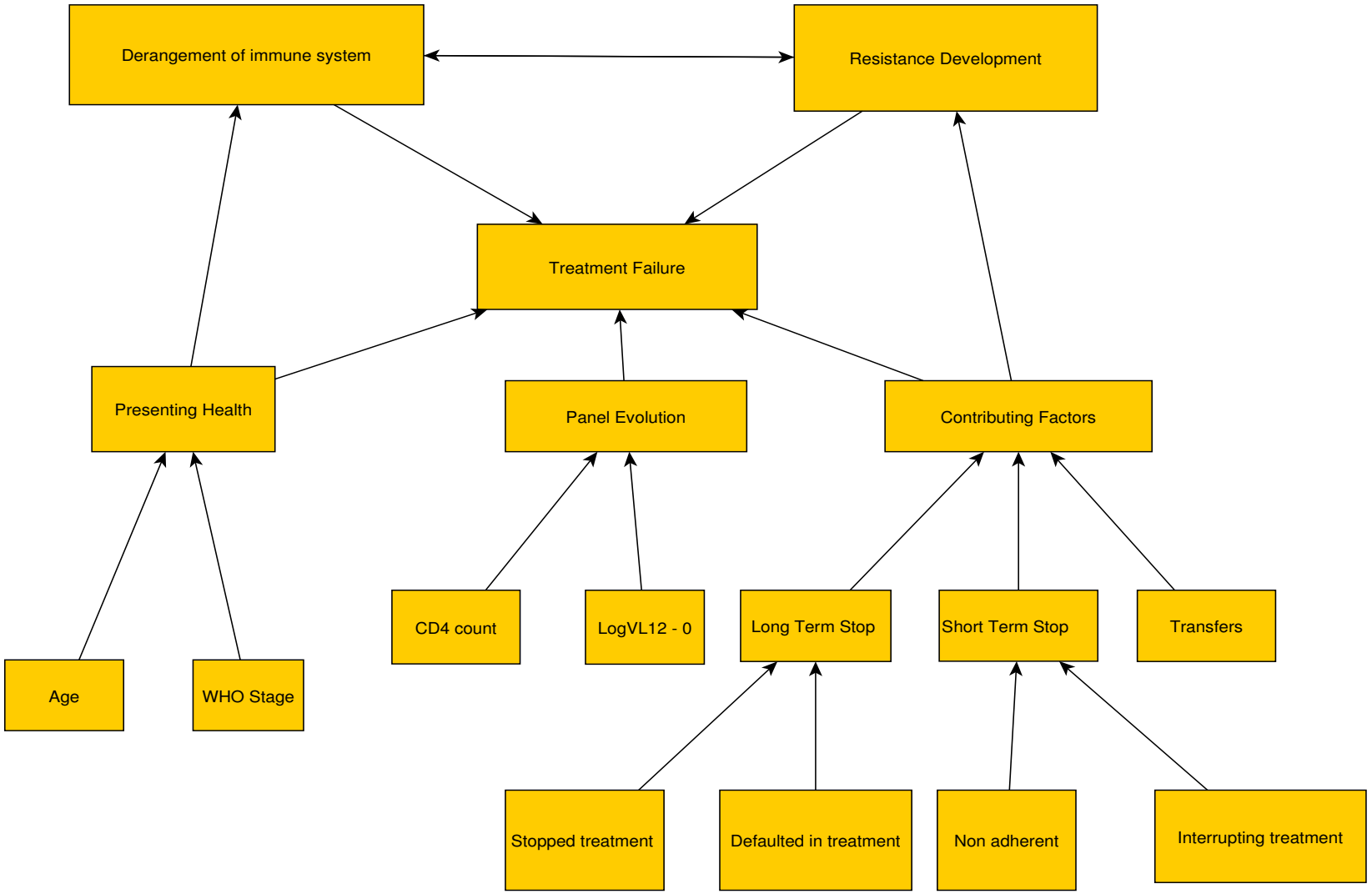


Figure 13: Final DAG representation of the causal hypothesis

The DAG (directed acyclic graph) in figure 13 is a causal hypothesis of treatment failure. The hypothetical causes include: (1) presenting health conditions such as age and WHO stage, (2) panel evolution such as the CD4 count which is a repeated measure over time and LogVL12-0 which is the difference in the log viral load at the 12month time point and baseline, (3) contributing factors such as long term stoppage of treatment which is stopping treatment completely or/and defaulting in treatment, short term stoppage of treatment which is non-adhering to treatment regimen or/and interrupting treatment and transfers from other health institutions. The contributing factors could lead to development of resistance while the presenting health conditions could lead to derangement of immune system. All of these factors can eventually result in treatment failure.

CHAPTER 4: DISCUSSION

This study specifically attempted to assess variables that are significantly associated with an increased risk of treatment failure. Identifying factors that are associated with an increased risk of treatment failure is advantageous as these can be used to put measures in place to try and delay or prevent the development of failure or its progression.

4.1 The proportion of patients that experienced treatment failure

In this study, a total number of 333 patients (38.28 %) experienced treatment failure throughout the study period. There were 437 instances of treatment failure which means that some patients failed treatment more than once. There were more cases of immunological failure (89.70 %) than virological failure (13.73 %). The proportion of patients that failed treatment in this study is higher compared to a study conducted Massachusetts General Hospital HIV outpatient clinic by Robbins et al in which 27.2 % of the patients in clinic population had treatment failure over a 2-year period[18] but lower than the proportion of patients that failed treatment in a study conducted by Barth et al in Ndlovu Medical Centre, Limpopo, South Africa which found that 46 % of the patients experienced treatment failure[65].

There was a decrease in the total number of patients that failed treatment from 102(11.72 %) at 12 month time point to the least number of failures – 37 patients (4.49 %) - at the 24 month time point. Subsequently this increased gradually through the 36 month (6.93 %), 48 month (12.27 %) to a maximum of 140 patients (17.01 %) at the 60 month time point.

4.2 Virological Failure

Virological failure was experienced by a small proportion of the study population: 45 patients (5.17 %) at 12 months, 14patients (1.70 %) at 24 month, only 1 patient (0.12 %) at 48 month and no one failed virologically at the 36 and 60 month time points. Suppression of viremia was achieved in most of the patients by the 36month as only one patient failed virologically from the 36month to 60month time point. A study conducted by Barth et al in South Africa found that 16(117/735) of the patients with

more than 3 months follow up experienced virological failure[65]. The EuroSIDA study in Europe found that at 48weeks 19 % of the patients from southern Europe experienced virological failure[64].

4.3 Immunological Failure

307 of the 870 patients (35.29 %) failed treatment immunologically, that is, 92 % of the total number of patients that failed treatment overall. The study by Dragsted et al found that 23 % of the study population (550/2347) experienced immunological failure (CD4+ count less than or equal to the pre-HAART value) within 6 – 12 months of initiation of HAART[66]. A Ghanaian study which monitored patients starting ART for 3 years found 13 % of the patients to have experienced immunological failure (Immunological failure was defined on the basis of WHO guidelines as a persistent CD4 cell count <100 cells/mm$^{>3}$, a CD4 cell count less than the pre-treatment level, or a CD4 cell count <50 % of the peak treatment level)[67]. These proportions are much lower than the proportion of patients that failed immunologically in this study and possibly due to differing definitions of immunological failure used in the different studies.

Immunological failure decreased to the lowest at the 24 month time point and then increased gradually from the 36 month up to a maximum at the 60 month time point. In view of this and the fact that viral suppression was achieved by most patients at the 36 month time point, it is possible that some patients experienced discordant responses, in which the HIV RNA plasma level is below the limit of detection but the CD4 cell count response is blunted[28]. A blunted CD4 response despite suppression of viral replication has often been attributed to host characteristics, particularly older age, lower pre-treatment CD4 count, drug toxicity and genetic variability as a possible modulator of immunological recovery [11, 28].

We were unable to demonstrate an association between immunological failure and age. However, there was a significant association between treatment failure and age.

4.4 Rates of change of CD4 count and Viral Load overtime

The mean rate of change of CD4 count decreased over time and even declined to a negative value at the 60 month time point. This indicates that the mean rate of change of CD4 count declined over time instead of increasing as expected suggesting that immune recovery became suboptimal over time.

Viral suppression occurred only in the first 3 years of treatment as shown by the negative slopes of the mean rate of change of log viral load but significant viral suppression occurred only within the first year of treatment. There was no significant difference in the mean rate of change from the second year to the fifth year of treatment indicating that the level of viral suppression within the first year of treatment is very important to the success of the therapy.

4.5 Determinants of treatment failure

When considering the overall results of this study, of the 333 adult patients who experienced treatment failure at the hospital, 56 patients (18.18 %) missed more than 3 doses according to self-report, 65 patients (22.41 %) stopped ARVs temporarily (usually due to toxicity) and subsequently restarted, 12 patients (3.91 %) stopped ARVs permanently, 67 patients (21.61 %) did not come back to the clinic for 3 months or longer, i.e. defaulted in treatment. Of the 333 patients who failed treatment, 31.61 % and 22.90 % stopped treatment in the short term or long term respectively. These figures are of concern in the light of developing resistance in the patients that take ARVs.

This study showed that adherence was one of the factors that strongly determine the occurrence of treatment failure. In the univariate comparisons, non-adherence, treatment interruption and defaulting were significantly different between the failure and non-failure groups. In the multivariate analysis both the STSTOP and LTSTOP were statistically significant. This shows that adherence to treatment and compliance to follow up appointments is crucial to the success of ARV treatment. In their study Patterson et al (2000) showed that adherence of 95 % was significantly associated

with successful virological outcome (viral loads below 400 copies/ml). Selamawit in their study in Addis Abba showed that a higher rate of poor adherence episodes is significantly linked with an increased hazard for treatment failure.

It is well recognized that complete suppression of viral replication is critical for long term durability of antiretroviral therapy and this is achieved only in optimal adherence. Suboptimal adherence is a primary factor responsible for the emergence of resistant strains of the HI virus which in turn results in treatment failure, reduction of future treatment options and potential transmission of drug resistant virus in the population[25, 35, 37].

Almost 22 % of the population that failed treatment missed their appointment dates for 3months or longer. Missing a follow up date at the hospital translates to the patient not getting the next supply of medication, hence the patient defaults treatment. This raises concern in terms of their adherence [13]. A high rate of missed appointment days has also been shown to be significantly associated with a high risk of treatment failure by Gregory et al[17]. One or more missed visits in the prior year, which could be another surrogate for treatment adherence, is an important predictor of treatment failure[18]. Consistent attendance at medical appointments plays a central role in both prolonging life and enhancing quality of life for persons living with HIV/AIDS[13].

This study showed that interruption of treatment, whether in the short term or long term, was an indicator of treatment failure even if the interruption was a clinical intervention as in the case of toxicity. ART interruption is problematic for patients treated with combination regimens of NNRTIs and NRTIs. This is due to the long half-life of NNRTIs and the comparatively short half- life of NRTIs. Patients who experience interruption of these regimens are exposed to functional mono-therapy with NNRTIs which then leads to development of drug resistance due to the low genetic barrier of these regimens[42]. The genetic barrier is the number of viral mutations required to overcome the drug-selective pressure and is an important factor for the development of drug resistance. High genetic barrier drugs such as the protease inhibitors require the accumulation of several mutations to overcome the drug-selective pressure while for the low genetic barriers such as the NNRTIs,

development of resistance can occur after a single mutation. Development of resistance due to the long half- life of NNRTIs can be countered by covering the 'tail' of the NNRTI – e.g. by giving a protease inhibitor for 1 week after stopping the regimen - but this is often neglected in practice.

In this study, the change in the viral load between baseline and 12months (LOGVLBL-12) was significantly associated with treatment failure. The multivariate regression demonstrated a strong negative association between treatment failure and the change in log viral load at 12months from baseline. This means that the patients who had a slower reduction in viral load (slower rate of viral suppression) within the first 12months after initiation of therapy, were at a higher risk of treatment failure. This is consistent with the studies by Thiebaut et al in a large multi-risk cohort of HIV-1 patients which indicated that sustained HIV RNA over 3.7log10 copies/ml (5000 copies/ml) between 4 and 12months after initiation of antiretroviral treatment is a major prognostic factor of clinical disease progression[39].

This study also showed that CD4 count was a predictor of treatment failure, in that low baseline CD4 was associated with treatment failure. The CD4 count was significant in both the univariate comparisons and the xtgee regression, though not significant in the random effect logistic regression xtlogit, presumably due to the inability to specify the autoregressive covariance structure. Owing to potential loss of information, CD4 count was not stratified into categories in this study. The significance of CD4 count was shared with the EuroSIDA[46]and the Thailand[43] studies which have found baseline CD4 level of less than 100 to be significantly associated with risk of developing treatment failure.

A study by Robbins et al indicated that a low CD4 count at the beginning of the observation period was associated with an increased risk of treatment failure over the following 2 years [18]. This result is contrary to the study by Phillips et al that found that there was no strong evidence that the lower CD4 cell counts and higher viral loads at baseline are associated with poorer virological outcome of ART[3]. However their study examined only virological failure, while this in study both virological failure and immunological failure were examined.

A significant association between the treatment regimen and treatment failure was seen in the multivariate regression. This study did not look at the specific treatment combination that was associated with treatment failure. Several published randomized clinical trials have shown that different antiretroviral regimens have different capacities to decrease the plasma viral load and raise the CD4 cell count thus leading to different clinical efficacy[15]. This may be due to the differing mechanisms of action and genetic barriers. A study from India demonstrated a significant association between treatment failure and Efavirenz based regimens [38]. Several factors, such as greater efficacy of new antiretroviral drugs, together with simplified dosing and less drug toxicity that might increase adherence could reduce the rate of treatment failure in patients receiving ART.

Another predictor of treatment failure in the multivariate regression was age. This study showed that the older the patients are, the less likely they were to fail treatment (coefficient ratio = -0.0517). This might be attributed to the fact that older people tended to be more responsible with their health. However, the data was skew towards older people, which may also have impacted the results. This is contrary to Gutierrez et al findings that an older age at initiation of HAART predicts an unfavourable outcome in patients[15] and other studies which have shown that younger age appears to be associated with a more rapid CD4 cell count increase[49]and older age has often been associated with a blunted CD4 response despite suppression of viral replication[28]. It has been hypothesized that the magnitude of immune restoration is dependent on thymus activity, which decreased with age[45.] A study by Fatkenheuer et al however found that age had no influence on treatment failure[16].

Baseline WHO stage was shown to be a determinant of treatment failure. Although this variable was not significant in the univariate comparisons, the regression model showed a significant association between WHO stage and treatment failure. Patients who commenced treatment at an advance stage of the disease (WHO stage 3 or 4) were at a lower risk of failing treatment (coefficient ratio = -0.4175).By examination of

the normality of this variable, most patients presented with advanced disease (stage 3 or 4 disease) and this right skewness may have influenced the results.

Another determinant of treatment failure was when patients were transferred from other treatment centres. This was significant in the multivariate regression. This might be as expected in that the majority of the patients were transferred from other treatment centres due to their not responding positively to treatment, or they may already have developed resistance to some of the medication.TDH offered special clinical services not routinely available in the state sector, such as resistance testing in the patients failing at that time. Virological failure in previously treated patients is often associated with the development of reduced susceptibility to most or all treatment drugs[31]. This category of patients requires special clinical intervention to restore their failing immune system and achieve optimum response to treatment. The availability of trained experts in the management of HIV/AIDS disease in clinics and hospitals will reduce the number of cases that are transferred to other hospitals.

In this study more women failed treatment than men. However, women did compose the majority of the treatment population and no significant association was found between gender and treatment failure in the multivariate regression. There was no significant correlation between place of residence, employment status and treatment failure. Despite this, it is worth noting that most of the patients who failed treatment were from the Mamelodi Township and 74 % were unemployed. Since most of these patients have to travel to get to the hospital for their monthly appointments, unavailability of transport fare may explain why many defaulted on their appointments at least once. Socioeconomic status is indeed a factor that needs to be addressed when looking at improving patient compliance with appointment schedules, which in turn has an impact on treatment failure[13].

4.6 LIMITATIONS

Being a record review, only information that was collected at the time the records were established could be captured. The information contained in the medical records is only as accurate as the person/s that entered it into the spread sheet.

Similarly, this study was based on the availability of laboratory results at periodic intervals; hence unavailability of these results and information on the variables of interest in the hospital record would ultimately impact the results. Missing data for some patients would have had an impact on the results.

The adherence status of the patient was based on self-report and only one measurement on adherence was used throughout the study period. Adherence status was recorded haphazardly and adherence at each time point was for the most part not updated in the patient record. Since adherence was self-reported, patients may have stopped therapy without it being reported or may have delayed initiation of HAART after prescription.

It was not possible to determine clinical failure in this study since the WHO stage was not routinely collected at each time point in the hospital record. Clinical failure is however such a late manifestation of treatment failure, often lagging behind the development of virological failure by a few years. It is consequently likely that it would not have provided additional information regarding the prediction of treatment failure.

Finally, this study was not designed to evaluate psychological criteria in terms of patient perceptions regarding the reasons for treatment failure, as these details were not available in the hospital record.

CHAPTER 5: CONCLUSION AND RECOMMENDATIONS

More than one–third of the patients receiving treatment in TDH failed treatment within the 5year study period. The regression results showed that the determinants of treatment failure were age, WHO stage, transfer from other institutions, short term stoppage of treatment, long term stoppage of treatment, CD4 cell count and the level of viral suppression within the first year of treatment (LOGVLBL-12).

The causes of treatment failure include the emergence of drug resistance[73] – most commonly due to low plasma levels of the ARVs secondary to non-adherence to medication and drug interactions - and derangement of the immune system. Since drug resistance emerges very early during treatment failure, the viral load should be monitored frequently in the first year of treatment. For this reason, the US guidelines[71] recommend measuring plasma viral load before initiation of therapy and preferably within 2–4 weeks, and not more than 8 weeks, after treatment initiation or after treatment modification. Repeat viral load measurement should be performed at 4–8-week intervals until the level falls below the assay's limit of detection. In this context, if the viral load response after initiation of treatment is slower than expected, a phenotypic susceptibility testing may play an important role in predicting treatment failure[31]. Due to cost constraints, the South African guidelines unfortunately no longer recommend a baseline VL and advise the first VL measurement to occur 6 months after the initiation of therapy. This strategy will likely miss patients at risk of developing treatment failure.

Patients interrupt or stop treatment for a number of reasons, emphasizing the fact that HAART regimens are complex and are often associated with side effects. Response to therapy has been shown to be highly dependent on adherence to therapy; hence a closer look into the reasons why patients chose to stop or interrupt therapy such as the need for more social support or information could help to improve adherence and clinical outcome.

Healthcare workers should also be aware of the need to cover the “tail” of the NNRTI-based regimens in order to prevent the development of drug resistance after ART is interrupted for reasons of toxicity.

The results from this study reinforce the need for identifying high risk patients earlier in treatment and to implement strategies that might strengthen adherence to treatment. Further studies that investigate the efficacy of intensive case management, adherence, support calls, treatment buddies, and directly observed therapy will assist clinicians in directing targeted disease management interventions to the high risk patients in order to improve treatment outcomes on ART.

Recommendations

- Documentation of patients’ information in the hospital file should be improved. All personnel involved in recording patients’ information must be trained to take a comprehensive history and document detailed information.
- Mechanisms should be implemented to identify patients at high risk of treatment failure early. One such strategy might be more intensive VL monitoring in the first 6 to 12 months of treatment. High risk patients should be flagged for closer monitoring.
- The determinants of treatment failure identified could be considered in combination and not in isolation.
- Adherence counselling might be strengthened throughout the whole system. Since adherence to the treatment regimen is one of the most important factors for a successful ART program, this cannot be over-emphasized. This should not be left to the counsellors and social workers alone but needs to be re-iterated by all personnel in the health facility.
- Patients must be empowered with information about their health condition. Patients should have a clear understanding of the importance of adherence and the implications of non-adherence such as drug resistance and cost implications, which will impact on successful ART management.
- Interruption of treatment by clinical intervention due to factors such as drug toxicity should be fashioned in a systematic manner so as to reduce the negative impact of interruption of treatment.

- The South African National HIV Treatment guideline stipulates that ART should be initiated at a CD4 cell count <200 cells/mm^3. The Minister of Health recently announced that this will be increased to a threshold of 350 cells/mm^3. Since treatment failure is associated with low CD4 count this step might help in counteracting the high rate of treatment failure. Patients are however still accessing treatment very late – the mean baseline CD4 count in this study was 109.57 cells/mm^3. Policy makers should now focus on educating patients to access treatment earlier.
- Closely linked to adherence is the regimen dosage. Availability of fixed dosage combination must be considered and expedited to improve adherence to treatment and hence reduce the failure rate.
- A viable system to identify patients who missed their appointment dates at the hospital could be established. Immediate and close follow-up of these patients will improve compliance to hospital visits and treatment regimen and ultimately lead to a successful clinical outcome.
- Special targeted disease management and intensive adherence counselling should be rendered to all high-risk patients including those transferred from other health institutions.
- Further *prospective* studies could be done to explore the underlying causes of non-adherence, treatment failure and occurrence of drug resistance.

REFERENCES

1. AIDSinfo (2008) HIV treatment regimen failure. http://aidsinfo.nih.gov (December 2008)
2. AIDSmed.com (2007) Understanding Drug Resistance. http://www.aidsmeds.com/articles/Resistance_7509.shtml (November 2010)
3. Phillips AN, Staszewski S, Weber R, et al. HIV Viral Load Response to Antiretroviral Therapy According to the Baseline CD4 Cell Count and Viral Load. JAMA, November 28, 2001—Vol 286, No. 20 : 2560-2567
4. Jaen A, Esteve A, Miro´ JM, et al. Determinants of HIV Progression and Assessment of the Optimal Time to Initiate Highly Active Antiretroviral Therapy PISCIS Cohort (Spain) J Acquir Immune Defic Syndr 2008;47:212–220
5. Cingolani A, Antinori A, Gabriella M, et al. Usefulness of monitoring HIV drug resistance and adherence in individuals failing Highly Active Antiretroviral Therapy: a randomized study (ARGENTA). AIDS(2002); 16(3):369-379
6. Bansi L, Sabin C, Delpech V, et al. Trends over calendar time in antiretroviral treatment success and failure in HIV clinic populations. HIV Medicine (2010) 11:432 – 438
7. Bartlett John, Cheever L, Johnson M et al.. A guide to primary care for people with HIV/AIDS(2004) . http://img.thebody.com/hrsa/pdfs/primary_care04.pdf.
8. Charles E. McCulloch. Repeated Measures And Hierarchical Models Lecture 2. http://www.epibiostat.ucsf.edu/dave/biostat209/Repeated measures 2.pdf
9. Continuing antiretroviral treatment. www.avert.org/arv-treatment.htm (2010)
10. Cornell M, Grimsrud A, Fairall L et al. Temporal changes in programme outcomes among adult patients initiating antiretroviral therapy across South Africa, 2002-2007. AIDS (2010, September) ;24(14): 2263–2270
11. Moore DM, Hogg RS, Yip B et al. Discordant Immunologic and Virologic Responses to Highly Active Antiretroviral Therapy Are Associated With Increased Mortality and Poor Adherence to Therapy. J Acquir Immune Defic Syndr 2005;40:288–293
12. Department of Health. The South African Antiretroviral treatment Guidelines 2010, South African National AIDS Council. http://www.doh.gov.za (2010).

13. Emmanuel Kgotso Ncholo. Review of patient follow-up mechanisms in the two Ekurhuleni Metropolitan Hospitals providing Antiretroviral treatment. (2009) http://wiredspace.wits.ac.za/handle/10539/7654
14. Steyerberg EW, Eijkemans MJ, Harrell FE, Habbema JD. Prognostic Modeling with Logistic Regression Analysis: In Search of a Sensible Strategy in Small Data Sets. Med Decis Making 2001; 21(1): 45-56
15. Gutierrez F, Padilla S, Masia M et al. Clinical Outcome of HIV-Infected Patients with Sustained Virologic Response to Antiretroviral Therapy: Long-Term Follow-Up of a Multicenter Cohort. PLoS ONE (2006) 1(1): e89. www.plosone.org/article/info:doi%2F10.1371%2Fjournal.pone.0000089
16. Fätkenheuer G, Theisen A, Rockstroh J et al. Virological treatment failure of protease inhibitor therapy in an unselected cohort of HIV-infected Patients. AIDS (1997) ;11(14):F113–F116
17. Gregory M. Lucas, Richard E. Chaisson, Richard D. Moore. Highly Active Antiretroviral Therapy in a Large Urban Clinic: Risk Factors for Virologic Failure and Adverse Drug Reactions. Ann Intern Med.1999; 131:81-87.
18. Gregory K. Robbins, Brock Daniels, Hui Zheng et al. Predictors of antiretroviral treatment failure in an urban HIV clinic. Journ.Acquired Immune Defic Syndr. 2007; 44(1): 30-37.
19. Gregory K. Robbins, Kristin L. Johnson, Yuchiao Chang et al. Predicting Virologic Failure in an HIV Clinic. Clin Infect Dis. 2010 March 1; 50(5): 779–786.
20. Government of South Africa (2007, 12th March) 'HIV and AIDS and STI Strategic Plan for South Africa, 2007-2011'. http://www.safaids.net
21. Impact of malnutrition on HIV treatment failure in resource-limited settings. John Hopkins centre for global health. http://research.hopkinsglobalhealth.org
22. Basenero A, Castelnuovo B, Birabwa E, et al. Inadequacy of clinical and immunological criteria in identifying virologic failure of 1st line ART the Ugandan experience. http://www.ias2007.org (2007)

23. Ioannidis JP, Cappelleri JC, Lau J, et al**.** Predictive value of viral load measurements in asymptomatic untreated hiv-1 infection: a mathematical model. AIDS *(*MAR 1996)**;** 10**(**3):255-262.
24. James Learned. Why HIV Drug Resistance Matters: An Overview. Positive Aware (2005); 16(5):26-30.
25. Goldman JD, Cantrell RA, Mulenga LB et al et al; Simple Adherence Assessments to Predict Virologic Failure among HIV-Infected Adults with Discordant Immunologic and Clinical Responses to Antiretroviral Therapy. AIDS Research and Human Retroviruses. August 2008, 24(8): 1031-1035.
26. Luc P and Amalio T. HIV treatment failure: Testing for HIV resistance in clinical practice. Science (1998); 280(5371): 1871
27. Mariel M Finucane, Jeffrey H Samet and Nicholas J Horton. Translation methods in biostatistics: linear mixed effect regression models of alcohol consumption and HIV disease progression overtime. Epidemologic Perspectives and Innovations 2007,4:8 www.epi-perspectives.com/content/4/1/8
28. Mauro Schechter and Suely Hiromi Tuboi. Discordant immunological and virological responses to antiretroviral therapy. Journal of Antimicrobial Chemotherapy (2006); 58(3): 506–510
29. Petersen ML, Mark J. van der Laan, Napravnik S et al. Long-term Consequences of the Delay Between Virologic Failure of Highly Active Antiretroviral Therapy and Regimen Modification. AIDS (2008); 22:2097–2106
30. Motsoaledi, A (2011, 31st May) 'How we're re-engineering the health system - Motsoaledi: Health Budget Vote Policy Speech presented at the National Assembly' politicsweb.co.za
31. Parkin NT, Deeks SG, Wrin MT et al. Loss of antiretroviral drug susceptibility at low viral load during early virological failure in treatment-experienced patients. AIDS 2000; 14:2877-2887
32. Paterson DL, Swindells S, Mohr J et al. Adherence to Protease inhibitor therapy and outcome in patients with HIV infection. Intern Med (July 4 2000); 133: 21-30.

33. Palella FJ, Delaney KM, Moorman AC et al. Declining morbidity and mortality among patients with advanced human immunodeficiency virus infection. N. Engl. J. Med. (1998); 338 (13): 853–860
34. Paredes R, Mocroft A, Kirk O et al. Predictors of virological success and ensuing failure in HIV-positive patients starting Highly Active Antiretroviral Therapy in Europe. Arch Intern Med. (2000) 160:1123-1132
35. Patel AK, Patel KK. Future implications: Compliance and failure with antiretroviral treatment. J Postgrad Med (2006); 52: 197-200
36. Paul S Albert. Tutorial in biostatistics: Longitudinal data analysis (repeated measures) in clinical trials. Statist. Med. (1999); 18: 1707-1732
37. Raboud JM, Harris M, Rae S et al. Impact of adherence on duration of virological suppression among patients receiving combination antiretroviral therapy. HIV Med 2002; 3:118-124.
38. Sikhamani R, Lakshmanan J, Sundaraj V. Predictors of failure of first-line antiretroviral therapy in HIV-infected adults: Indian experience. AIDS. 2007; 21(S4):S47–S53
39. Thiebaut R, Morlat P, Jacqmin-Gadda H et al. Clinical progression of HIV-1 infection according to the viral response during the first year of antiretroviral treatment. AIDS (2000); 14: 971-978
40. Rosen S, Ketlhapile M, Sanne I, De Silva, M.B. Cost to Patients of Obtaining Treatment for HIV/AIDS in South Africa. South African Medical Journal (2007); 97 (10)**:**524-529
41. SANAC (2010, February) 'The national HIV counselling and testing campaign'. South Africa National AIDS Council (http://www.sanac.org.za)
42. Selamawit Ejigu. First line antiretroviral treatment failure and factors associated with it in Addis Ababa. http://etd.aau.edu.et (2009)
43. Srasuebkul P, Ungsedhapand C, Ruxrungtham K, et al. Predictive factors for immunological and virological endpoints in Thai patients receiving combination antiretroviral treatment. HIV Medicine. 2007; 8: 46–54
44. Little SJ, Holte S, Routy JP et al. Antiretroviral-drug resistance among patients recently infected with HIV. The New England Journal of Medicine (2002); 347: 385-395

45. Teixeira L, Valdez H, McCune J et al. Poor CD4 T cell restoration after suppression of HIV-1 replication may reflect lower thymic function. AIDS 2001; 15(14) 1749-1756.
46. Dragsted UB, Mocroft A, Vella S et al. Predictors of Immunological Failure after Initial Response to Highly Active Antiretroviral Therapy in HIV-1–Infected Adults: A EuroSIDA Study. Journal of Infectious Diseases (2004); 190: 148-155
47. UNAIDS report www.unaids.org (2011)
48. UNAIDS reports on global AIDS epidemic.www.unaids.org (2009, November)
49. Viard J, Mocroft A, Chiesi A et al. Influence of age on CD4 cell recovery in HIV-infected patients receiving HAART: evidence from the EuroSIDA study. Journal of Infectious Diseases (2001); 183:1290-1294.
50. Walmsley S. Update on antiretroviral treatment failure and the management of treatment-experienced HIV-infected patients. (2006) www.thebody.com/content/art42267.html
51. WHO. Antiretroviral therapy for HIV infections in adults and adolescents. Recommendations for a public health approach. www.who.int/hiv/pub/guidelines/artadultguidelines.pdf
52. WHO/UNAIDS/UNICEF 'Towards Universal Access: Scaling up priority HIV/AIDS interventions in the health sector' http://whqlibdoc.who.int/publications/2010/9789241500395_eng.pdf. (2010)
53. Yazdan Y. Multidrug resistance: a clinical approach. Current Opinion HIV in AIDS (2009); 4: 499-506
54. Zalar A, Figueroa MI, Ruibal-Ares B et al. Macrophage HIV-1 infection in duodenal tissue of patients on long term HAART. Antiviral Research (2010) 87: 269-271
55. Hirsch MS, Conway B, D'Aquila RT, et al. Antiretroviral drug resistance testing in adults with HIV infection: Implications for clinical management. JAMA. 1998; 279: 1984-1991.
56. JA. Bartlett. Addressing the challenges of Adherence. JAIDS. (2002); 29: S2 – S10.
57. Henrard DR, Phillips JF, Muenz LR, et al. Natural history of HIV-1 cell-free viremia. JAMA (1995). 274:554-558

58. Choi S, Lagakos SW, Schoolev RT et al. CD4+ lymphocytes are an incomplete surrogate maker for clinical progression in persons with asymptomatic HIV infection taking zidovudine. Ann Intern Med (1993); 118: 674-680.
59. Hooper CJ, Coombs RW, Boulin PD et al. Increase in CD4 cell count are associated with decrease in HIV-1 viral load in patients changing antiretroviral regimens. 35th Interscience Conference on Antimicrobial Agents and Chemotherapy. San Francisco. September 1995 [abstract 1152].
60. Clotet B, Ruiz L, Ibanez A et al. Viral load (VL) in plasma as a predictor in HIV- 1 infected patients on zidovudine therapy to which ddC is added. 35th Interscience Conference on Antimicrobial Agents and Chemotherapy. San Francisco. September 1995 [abstract 1153].
61. Mocroft A, Phillip AN, Ledergerber B et al. Estimated average annual rate of change of CD4 T-cell count in patients on combination ART. Antiviral Therapy (2010); 15: 563-570
62. Hogg RS, Yip B, Chan KJ et al. Rates of disease progression by baseline CD4 cell count and viral load after initiating triple-drug therapy. JAMA (November 28, 2001); 286 (20): 2568-2577
63. Swihart BJ, Caffo B, James BD et al Lasagna plots: A saucy alternative to spaghetti plots. (October 2009) www.bepress.com/cgi/viewcontent.cgi?article=1141&context=jhubiostat
64. Mocroft A, Miller V, Chiesi A et al. Virological failure among patients on HAART from across Europe: results from the EuroSIDA study. Antiviral Therapy (June 2000); 5: 107 - 112
65. Barth RE, Tempelman HA, Moraba R et al. Long-Term Outcome of an HIV-Treatment Programme in Rural Africa: Viral Suppression despite Early Mortality. AIDS Research and Treatment. Volume 2011, Article ID 434375, 5 pages. doi:10.1155/2011/434375
66. Dragsted UB, Mocroft A, Vella S et al. Predictors of ImmunologicalFailure after Initial Response to Highly Active Antiretroviral Therapy in HIV-1-Infected Adults. Journal of Infectious Diseases (July 1, 2004); 190(1): 148-155.
67. Collini P, Schwab U, Sarfo S et al. Sustained Immunological Responses to Highly Active Antiretroviral Therapy at 36 Months in a Ghanaian HIV Cohort. Clinical Infectious Diseases (April 1, 2009). 48(7): 988-991.

68. UNAIDS (2010) 'UNAIDS report on the global AIDS epidemic' . http://www.unaids.org (2010)
69. Wikipedia -WHO disease staging system for HIV infection and disease. http://en.wikipedia.org
70. Ian Shrier and Robert W Platt. Reducing bias through directed acyclic graphs. Bmc medical research methodology (2008); 8:70. Http://www.biomedcentral.com
71. Guidelines for the Use of Antiretroviral Agents in HIV-1-Infected Adults and Adolescents (January 10, 2011). Developed by the DHHS Panel on Antiretroviral Guidelines for Adults and Adolescents – A Working Group of the Office of AIDS Research Advisory Council (OARAC). P9
72. Schouten JT, HIV drug resistance and the other causes of treatment failure. Step Perspective. 1997 Winter:9(3): 5-8

APPENDICES

Appendix 1: Study tool.

STUDY NO:

HOSPITAL NUMBER:

A	**DEMOGRAPHICS:**					
	Age					
	Gender		Male		Female	
	Place of residence					
B	**IMMUNOLOGICAL STATUS**					
	CD4 Count at Baseline	CD4 Count at 12 months	CD4 Count at 24 month	CD4 Count at 36month	CD4 Count at 48month	CD4 Count at 60month
C	**HISTORY OF VIRAL LOAD (VL)**					
	VL at Baseline	VL at 12 months	VL at 24months	VL at 36month	VL at 48month	VL at 60month
D	**ARV TREATMENT HISTORY:**					
	ARV used before starting HAART:					
	Number of treatment regimen exposed to					
	ARV treatment exposed to at:					
	Baseline	12 months	24months	36month	48month	60month
E	**WHO STAGING :**					
	Baseline	12 months	24months	36month	48month	60month
F	**CONCOMITANT DISEASE:**					
	Baseline	12 months	24months	36month	48month	60month
G	**ADHERENCE COUNSELLING (YES OR NO)**					
	Baseline	12 months	24months	36month	48month	60month

Appendix 2: World Health Organization (WHO) Clinical Staging of HIV/AIDS For Adults and Adolescents (2005)

Primary HIV infection

- Asymptomatic
- Acute retroviral syndrome

Clinical stage 1

- Asymptomatic
- Persistent generalized lymphadenopathy

Clinical stage 2

- Moderate and unexplained weight loss (<10 % of presumed or measured body weight)
- Recurrent respiratory tract infections (such as sinusitis, bronchitis, otitis media, pharyngitis)
- Herpes zoster
- Recurrent oral ulcerations
- Papular pruritic eruptions
- Angular cheilitis
- Seborrhoeic dermatitis
- Fungal finger nail infections

Clinical stage 3

Conditions where a presumptive diagnosis can be made on the basis of clinical signs or simple investigations

- Unexplained chronic diarrhoea for longer than one month
- Unexplained persistent fever (intermittent or constant for longer than one month)
- Severe weight loss (>10 % of presumed or measured body weight)
- Oral candidiasis
- Oral hairy leukoplakia

- Pulmonary tuberculosis (TB) diagnosed in last two years
- Severe presumed bacterial infections (e.g. pneumonia, empyema, meningitis, bacteraemia, pyomyositis, bone or joint infection)
- Acute necrotizing ulcerative stomatitis, gingivitis or periodontitis

Conditions where confirmatory diagnostic testing is necessary

- Unexplained anaemia (< 80 g/l), and or neutropenia (<500/µl) and or thrombocytopenia (<50 000/ µl) for more than one month

Clinical stage 4

Conditions where a presumptive diagnosis can be made on the basis of clinical signs or simple investigations

- HIV wasting syndrome
- Pneumocystis pneumonia
- Recurrent severe or radiological bacterial pneumonia
- Chronic herpes simplex infection (orolabial, genital or anorectal of more than one month's duration)
- Oesophageal candidiasis
- Extrapulmonary Tuberculosis
- Kaposi's sarcoma
- Central nervous system toxoplasmosis
- HIV encephalopathy

Conditions where confirmatory diagnostic testing is necessary

- Extrapulmonary cryptococcosis including meningitis
- Disseminated non-tuberculous mycobacteria infection
- Progressive multifocal leukoencephalopathy
- Candida of trachea, bronchi or lungs
- Cryptosporidiosis
- Isosporiasis
- Visceral herpes simplex infection

- Cytomegalovirus (CMV) infection (retinitis or of an organ other than liver, spleen or lymph nodes)
- Any disseminated mycosis (e.g. histoplasmosis, coccidiomycosis, penicilliosis)
- Recurrent non-typhoidal salmonella septicaemia
- Lymphoma (cerebral or B cell non-Hodgkin)
- Invasive cervical carcinoma
- Visceral leishmaniasis

Appendix 3: University of the Witwatersrand Ethics clearance

UNIVERSITY OF THE WITWATERSRAND, JOHANNESBURG
Division of the Deputy Registrar (Research)

HUMAN RESEARCH ETHICS COMMITTEE (MEDICAL)
R14/49 Ms Temitope O Sokoya

CLEARANCE CERTIFICATE	**M10928**
PROJECT	Occurrence and determinants of treatment failure in antiretroviral therapy at Tshwane distric Hospital.
INVESTIGATORS	Ms Temitope O Sokoya.
DEPARTMENT	Department of Pharmacy & Pharmacology
DATE CONSIDERED	01/10/2010
DECISION OF THE COMMITTEE*	Approved unconditonally

Unless otherwise specified this ethical clearance is valid for 5 years and may be renewed upon application.

DATE 01/10/2010 **CHAIRPERSON** .....
(Professor PE Cleaton-Jones)

*Guidelines for written 'informed consent' attached where applicable
cc: Supervisor : Prof BM Gundidza

DECLARATION OF INVESTIGATOR(S)

To be completed in duplicate and **ONE COPY** returned to the Secretary at Room 10004, 10th Floor, Senate House, University.
I/We fully understand the conditions under which I am/we are authorized to carry out the abovementioned research and I/we guarantee to ensure compliance with these conditions. Should any departure to be contemplated from the research procedure as approved I/we undertake to resubmit the protocol to the Committee. **I agree to a completion of a yearly progress report.**
PLEASE QUOTE THE PROTOCOL NUMBER IN ALL ENQUIRIES...

Appendix 4: Tshwane Metsweding Region Research Ethics clearance

health and social development
Department: Health and Social Development
GAUTENG PROVINCE

Umnyangowezempilo no Kuthuthukiswa Komphakathi
Lefapha la Maphelo le Tshebeletso le Ntshetsopele ya Sechaba
Department of Health and Social Development
Departement van Gesondheid en Maatskaplike Ontwikkeling

TSHWANE/METSWEDING REGION
Enquiries: Ms M Mosito
+27725602528
Email:Motlalepule.Mosito@gauteng.gov.za

TSHWANE METSWEDING REGION RESEARCH ETHICS COMMITTEE

CLEARANCE CERTIFICATE

Meeting: 02/2011

PROJECT NUMBER: TMREC 2011/05

PROJECT:

Title:	Occurrence and determinants of treatment failures in anti retroviral therapy at TDH
Researcher:	Temitope Sokoya
Supervisor:	Prof Gundidza
Co-supervisor:	Dr T Rossouw, Dr M Nieuwoudt
Department:	Pharmacy & Pharmacology (WITS)
Degree:	MSc

DECISION OF THE COMMITTEE

Approved

Date: 22 February 2011

.....
Dr K.E Letebele-Hartell
Chairperson Tshwane Metsweding Research Ethics Committee
Tshwane Metsweding Region

.....
Mr M Pitsi
Director: District Health Services Support
Tshwane Metsweding Region

NOTE: Resubmission of the protocol by researcher(s) is required if there is departure from the protocol procedure as approved by the committee.
ALL CORRESPONDANCE TO INCLUDE PROTOCOL NUMBER

Appendix 5: TDH letter of permission

JAN 2011

Department of Health and Social Development
Umnyango wezempilo no Kuthuthukiswa Komphakathi
Letapha la Maphelo le Tshebeletso le Ntshetsopele ya Sechaba
Departemente van Gesondheid en Maatskaplike Ontwikkeling

TSHWANE DISTRICT HOSPITAL

TO: **Dr Letebele**
Tshwane Metsweding Regional Office
District Health Research Ethics Committee (DHREC)

FROM: **Dr HM Mosoane: Senior Clinical Manager**
DATE: 02 | 02 | 2011

RE: PERMISSION TO CONDUCT RESEARCH AT TDH

In compliance with the GDoH Research Coordination Policy Guidelines, please find herewith research documents for authorization.

The research topic: Occurrence and determinants of treatment failures in Antiretroviral therapy at TDH

Researcher & Contact details: Temitope Jokoya 072642 0338

University: Wits MMED Student: YES / NO / Other Student MSc

Department of Health & Social Development Employee: (YES) / NO

Supervisor(s): Prof Gundidza

Institution & unit where research will be undertaken TDH

1. Full Copy of Proposal: (YES) / NO
2. Ethical approval from an institutional Research Ethics Committee: YES / (NO) Not available
   The student will e-mail it as requested.

(Recommended) / Not Recommended

pp

Dr Soe: Acting CEO Tshwane District Hospital

Permission granted / not granted

Chairperson DHREC

Chief Director Tshwane/Metsweding Region

1

Tshwane District Hospital
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