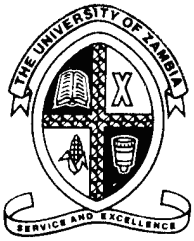


## The University of Zambia

## School of Medicine

**Department of Obstetrics and Gynaecology**

# A STUDY OF NEVIRAPINE TOXICITY IN HIV INFECTED PREGNANT WOMEN AT THE UNIVERSITY TEACHING HOSPITAL IN LUSAKA, ZAMBIA

**Dissertation submitted in partial fulfillment of the requirement and for the degree of Masters of Medicine in Obstetrics and Gynaecology**

Jessica Mulindwa
2012

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## ABBREVIATIONS

| | |
|---|---|
| ACTG | AIDS Clinical Trial Group |
| AIDC | Adult Infectious Disease Centre |
| AIDS | Acquired Immunodeficiency Syndrome |
| ART | Antiretroviral Therapy |
| AST | Aspartate Amino Transferase |
| ALT | Alanine Amino Transferase |
| CDC | Centers for Disease Control and Prevention |
| CIDRZ | Centre for Infectious Disease Research in Zambia |
| DHHS | United States Department of Health and Human Services |
| FBC | Full Blood Count |
| FDA | Food and Drug Agency |
| HAART | Highly Active Antiretroviral Therapy |
| LFT | Liver Function Test |
| MOH | Ministry of Health |
| NNRTI | Non Nucleoside Reverse Transcriptase Inhibitor |
| PI | Protease Inhibitor |
| PMTCT | Prevention of Mother to Child Transmission (of HIV/AIDS) |
| RPR | Rapid Plasma Reagin |
| US | United States |
| UTH | University Teaching Hospital |
| WHO | World Health Organization |
| ZEPRS | Zambia Electronic Perinatal Record system |

## ACKNOWLEDGEMENTS

I am deeply indebted to the patients who took part in this study, my supervisors Dr. Bellington Vwalika and Dr. Namwinga Chintu, my ICRS (International Clinical Research Scholars) Fogarty scholarship supervisor Dr. Benjamin Chi, my clinical supervisor at the Adult Infectious Disease Centre (AIDC) Dr. Elizabeth Stringer, the University Teaching Hospital department of Obstetrics and Gynaecology as well as the entire CIDRZ research team for their unwavering support to see to the successful completion of this study. I also remain grateful to Ms Elizabeth Sitali, the AIDC clinic co-coordinator for her meticulous organization of the AIDC clinic which allowed for collection of data with relative ease.

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Dr Jessica Mulindwa

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## DECLARATION

I hereby declare that this dissertation represents my own work and has not been presented even in part to any forum or University other than the University of Zambia and the CIDRZ research team.

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## APPROVAL

This dissertation of Dr Jessica Mulindwa is approved as partial fulfillment of the requirements for the award of the Master of Medicine in Obstetrics and Gynaecology by the University of Zambia

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   Date_____

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   Date_____

3. Name_____

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   Date_____

## ABSTRACT

### Background

The toxicity of nevirapine based regimen in pregnant women with CD4 counts greater than 250 cells/$mm^3$ is well documented. WHO as well as the United States Food and Drug Agency (US FDA) cautions use of nevirapine in pregnant women with CD4 counts greater than 250 cells/$mm^3$. The Zambian guidelines recommend initiating HAART for all HIV positive pregnant women with CD4 counts less than 350 cells/$mm^3$. Nevirapine based regimen is the recommended first line, even for CD4 counts between 250 to 350cell/$mm^3$. We compared incidence of nevirapine toxicity in pregnant HIV infected women commenced on a nevirapine based regimen in pregnancy with CD4 counts less than 250 cells/$mm^3$ to that of women with CD4 counts between 250 and 350 cells/$mm^3$.

### Methods

Longitudinal observational study with 2 arms; CD4 count below 250 cells/ $mm^3$, and CD4 count of 250-350 cells/ $mm^3$. Convenience sampling was used to enroll 146 patients. All HIV infected pregnant women with CD4 counts less than or equal to 350cells/$mm^3$ commenced on nevirapine based regimen in the current pregnancy were enrolled if they consented and they fulfilled the eligibility criteria. Women were allocated to the 2 arms based on their CD4 count. An interviewer administered questionnaire was used to collect demographic details at the recruitment stage. On subsequent visits, a check list was used to collect data.

SAS statistical package was used to analyze the data.

**Results**

The incidence of nevirapine toxicity Rash (at least grade 2 rash with or without mucous membrane involvement and/or at least grade 2 rise in Aspartate Amino Transferase) was found to be 0% in women with CD4 counts below 250cells/$mm^3$; it was 13.5% ($p$=0.005) in women with CD4 counts between 250 to 350cell/$mm^3$.

**Conclusions**

Though the study was powered to detect a 15% difference, the results of the study show that women with CD4 counts of 250 to 350cells/$mm^3$ are at substantial risk of nevirapine toxicity when commenced on nevirapine based HAART regimen in pregnancy.

# 1 INTRODUCTION

The use of Highly Active Antiretroviral Therapy (HAART) to prevent mother-to-child HIV transmission (PMTCT) has demonstrated to be effective (US Public Health Services Task Force 2005, Centers for Disease Control and Prevention 2006). This has been shown to be true also in developing countries even in the face of breastfeeding (Read et al 2005, Palombi et al 2007).

Data on safety of nevirapine in pregnancy has given rise to concern. Risk of toxicity has been shown to increase with CD4 counts greater than 250cells/$mm^3$ especially during pregnancy (Dieterich D et al 2001, Hitti J et al 2004, Jamisse L et al 2007).
The US Food and Drug Administration (FDA) as well as the World Health Organization advises use of nevirapine based regimen in women with CD4 counts greater than 250 with caution (FDA advisory-2005, WHO 2007).

Nevirapine has been widely used in pregnancy both as single-dose therapy during labour and as part of short-term and long-term highly active antiretroviral therapy. It is an effective and well tolerated non-nucleoside reverse transcriptase inhibitor (NNRTI) that is widely available to prevent HIV infection in resource-poor populations (US DHHS 2006, Marazzi et al 2006).

In developing countries, choice of highly active antiretroviral drugs in pregnancy for therapy or for PMTCT remains a challenge. Efavirenz is a non nucleoside reverse transcriptase inhibitor that has been associated with neural tube defects when used in the

first trimester. Niverapine based regimens have well documented cutaneous and hepatic toxicities when used in women with high CD4 counts. Nevirapine based regimen is inexpensive compared to both efavirenz and protease inhibitor based regimens (Sabbatani et al 2005). Protease inhibitor (PI) based regimens form the back-bone of second line therapy and are therefore not recommended as first line regimen.

The Zambian MOH 2007 ART protocol guidelines (as well as WHO 2006 ART guidelines in resource poor settings) recommend initiating HAART for all HIV-1 infected pregnant women with CD4 counts less than 350cells/$mm^3$. Nevirapine based regimen is the first line therapy for pregnant women with CD4 counts less than 250cells/$mm^3$. For women with CD4 counts between 250 and 350cells/$mm^3$, either nevirapine with frequent checks on liver function tests or efavirenz commenced after the $1^{st}$ trimester is recommended (The Zambian MOH 2007 ART protocol guidelines).

The study determined the incidence of nevirapine toxicity in HIV-1 infected women commenced on nevirapine based regimen (zidovudine/lamivudine/nevirapine or stavudine/lamivudine/nevirapine) in the current pregnancy with CD4 counts up to 350 cells/$mm^3$ at the University Teaching Hospital, Lusaka, Zambia. It compared the incidence of nevirapine toxicity (cutaneous and hepatic toxicity) in women with CD4 counts below 250cells/$mm^3$ to that of women with CD4 counts between 250 to 350cells/$mm^3$.The study also documented the prevalence of hepatitis B co-infection in the study population as this is an important confounder.

## 2 LITERATURE REVIEW

The effectiveness of highly active antiretroviral therapy during pregnancy for the purposes of treating as well as reducing the risk of the mother transmitting the virus to her baby is well established in developed countries (US Public Health Services Task Force 2005, Centers for Disease Control and Prevention 2006). Few studies done in developing countries have shown comparable efficacy even in the face of breastfeeding (Read et al 2005, Palombi et al 2007).

Safety data on use of nevirapine in pregnancy became an issue of concern following the AIDS Clinical Trials Group (ACTG) 1022 study where five out of seventeen (29.4%) pregnant women had nevirapine toxicity with one death from fulminant hepatitis (Hitti et al 2004).

However, most recommended regimens in developing countries are nevirapine based. This is because nevirapine is an inexpensive, effective and well tolerated non-nucleoside reverse transcriptase inhibitor (NNRTI) that is widely available to prevent HIV infection in resource-poor populations (US DHHS 2006, Marazzi et al 2006). It is non inferior to other non nucleoside reverse transcriptase inhibitors or protease inhibitors in its virological or immunological effects (Saag et al 2006, Ena et al 2008). Its pharmacokinetics is not significantly affected by pregnancy despite associated physiological changes, such that no dosage adjustments are required (Taylar 2000, Minochnick 2001). Nevirapine has been used in pregnancy for its efficacy, low pill burden, bioavailability and rapid transplacental transfer. It has been widely used both as

single-dose therapy during labour and as part of short-term, and long-term highly active antiretroviral therapy to minimize risk of vertical transmission of HIV.

In developing countries, choice of highly active antiretroviral drugs in pregnancy for therapy or for PMTCT remains a challenge. Antiretroviral drugs are either in groups B or C of the FDA (Food and Drug Administration) classification and all of them may cause toxicity. The United States Department of Health and Human Services (US DHHS) recommends maximum attention on a possible increase of nevirapine toxicity among HIV infected women with CD4 counts >250 especially during pregnancy (FDA advisory on nevirapine-2005). The US DHHS as well as WHO recommend that pregnant women with $CD4^+$ counts >250 cells/$mm^3$ should begin taking nevirapine only if the expected benefits clearly outweigh the risk of toxicity, although women who are tolerating nevirapine well when they enter pregnancy may continue therapy, regardless of their $CD4^+$ count (US DHHS ART guidelines 2006, WHO ART guidelines 2006). Efavirenz is a non-nucleoside reverse transcriptase inhibitor that has been associated with neural tube defects when used in the first trimester. Other non-nucleoside reverse transcriptase inhibitors are more expensive and/or are not available for use in public institutions. Protease inhibitor based regimens are not recommended for use as first line regimens as they form the backbone of second line therapy. Nevirapine is inexpensive compared to both efavirenz and protease inhibitor based regimens. A study done in Italy found that nevirapine had a 57% lower cost versus efavirenz, and even more difference versus protease inhibitors (Sabbatani et al 2005).

Skin complaints during normal pregnancy are common (Henson et al 2000, Black 2002). Their causes may include cholestasis, herpes gestationalis, pruritic urticarial papules and erythema nodosum. The commonest side effect associated with nevirapine is a maculopapular rash. In randomized controlled trials the risk of rash has been shown to be about 3% in women with CD4 counts<250 and about 13% in women with higher CD4 counts (Pollard et al 1998) and, risk of Steven-Johnson Syndrome (SJS) is 0.3% (Metry et al 2001).

Nevirapine also causes hepatitis, which may be asymptomatic or severe. Risk of nevirapine hepatotoxicity is about 3% in women with CD4 counts below 250 and about 11-13% in women with higher CD4 counts (Dieterich et al 2001, Martinez et al 2001). Life threatening complications of pregnancy such as acute fatty liver of pregnancy and the HELLP (haemolysis, elevated liver enzymes, low platelets) syndrome as well as hepatitis from nucleoside reverse transcriptase inhibitors can be confused with nevirapine hepatotoxicity (Glanville et al 2003).

Risk of nevirapine toxicity has been shown to be associated with higher CD4 counts when commencing therapy (Dieterich et al 2001, Jamisse et al 2007) and female gender (Bersoff-Matcha et al 2001). Higher baseline ALT/AST is associated with increased risk of toxicity (Dieterich et al 2001, Stern et al 2003)

HIV infected pregnant women co-infected with hepatitis B or C infection are at increased risk of developing Nevirapine hepatotoxicity (Dieterich et al 2001, Stern et al 2003, Joao

et al 2006). Being black is not a risk factor for developing nevirapine toxicity (Gonzalez-Garcia et al 2004, Natarajan et al 2007)

HIV-infected pregnant women exposed to nevirapine based HAART regimen for the first time during pregnancy are more likely to develop toxicity compared to those continuing nevirapine-based regimen (Natarajan et al 2007, Schalkwyk et al 2008). A prospective cohort study of pregnant HIV-1 infected women and non-pregnant HIV-1 infected women found that pregnancy was significantly associated with increased risk of hepatotoxocity in HIV-infected women (Ouyang et al 2008). Furthermore, women are more likely to commence HAART for either therapy or for prophylaxis when they are pregnant.

According to the 2007 Zambian ART guidelines (as well as WHO 2006 ART guidelines in resource-poor settings) all HIV infected pregnant women with CD4 counts less than 350cells/$mm^3$ should be started on HAART. Nevirapine based regimen being the preferred first line regimen.

The prevalence of HIV infection among women attending antenatal in clinics around Lusaka is about twenty two percent (22%). Prevalence can be as high as 38 % in some centers (Lusaka DHMT/CIDRZ Programmatic Data 2008). The 2007 Zambia Demographic and Health Survey Report estimates that one out of three women is HIV positive in areas worst hit. As Ministry of Health Zambia together with its cooperating partners rolls out the PMTCT program, more HIV infected women are likely to

commence nevirapine-based HAART regimen while pregnant. There is therefore need to evaluate the incidence of nevirapine toxicity in pregnant women presenting with CD4 counts between 250 to 350cells/$mm^3$ and compare it to the incidence of those starting therapy with CD4 counts below 250cell/$mm^3$. Studies evaluating nevirapine toxicity in an African population have not been well established.

## 3 STATEMENT OF THE PROBLEM

Nevirapine is an effective and well tolerated non-nucleoside reverse transcriptase inhibitor (NNRTI) that is widely available to prevent HIV infection in resource-poor populations. It is inexpensive compared to other non-nucleoside reverse transcriptase inhibitors and protease inhibitors.

Though concerns regarding use of nevirapine based regimens for CD4 counts > 250 are established, no studies have been done to evaluate the cut-off or the threshold CD4 for increased risk of nevirapine toxicity.

The 2007 Zambian ART guidelines recommend nevirapine based regimen as the preferred regimen for all HIV-1 infected pregnant women with CD4 counts below 350cells/$mm^3$. For women with CD4 counts between 250 and 350cell/$mm^3$, more frequent monitoring of the patient is recommended.

There is need to evaluate the incidence of nevirapine toxicity (hepatotoxicity and cutaneous toxicity) in pregnant women who are started on nevirapine based regimen with

CD4 counts below 350cells/mm$^3$. Is the incidence of toxicity higher in women with CD4 counts between 250 to 350cells/mm$^3$? Furthermore the prevalence of Hepatitis B co-infection in HIV positive pregnant women in Zambia has not been well documented. Pregnant HIV positive patients who are co-infected with Hepatitis B may show an increased tendency to hepatotoxicity.

As the Ministry of Health together with its development partners roll out the PMTCT program in Zambia, more women with CD4 counts below 350cells/mm$^3$ are likely to commence nevirapine based highly active antiretroviral therapy regimen in pregnancy.

## 4 HYPOTHESIS

HIV-infected women commenced on a nevirapine regimen in pregnancy with CD4 counts less than 250 cells/mm$^3$ are less likely to develop nevirapine toxicity compared to HIV-infected women commenced on nevirapine regimen in pregnancy with CD4 counts between 250 cells/mm$^3$ and 350 cells/mm$^3$.

### *General Objective*

The general objective of the study was to determine incidence of nevirapine toxicity in pregnant HIV infected women commenced on nevirapine-based regimen in the current pregnancy with CD4 counts up to 350 cells/mm$^3$ at the University Teaching Hospital, Lusaka, Zambia.

***Specific objectives***

1) To compare the incidence of nevirapine toxicity in HIV-1 infected women commenced on nevirapine based regimen in pregnancy with CD4 counts less than 250 cells/$mm^3$ to that of women with CD4 counts between 250 and 350 cells/ $mm^3$ at the University Teaching Hospital, Lusaka, Zambia.
2) To determine the prevalence of hepatitis B co-infection in the study population
3) To describe the social and demographic characteristics associated with nevirapine toxicity in the study population.

## 5 DESIGNS AND METHODS

### *5.1 Study design*

Longitudinal observational study with 2 arms

Group 1 (low CD4 count arm): HIV infected pregnant women with CD4 counts less than 250 cells/$mm^3$ commenced on nevirapine based regimen in pregnancy.

Group 2 (higher CD4 count arm): HIV infected pregnant women with CD4 counts between 250 and 350 cells/$mm^3$ commenced on nevirapine based regimen in pregnancy.

### *5.2 Study duration*

The study took about 11 months from inception

- Training study assistants-1 week
- Baseline data collection -8 months
- Additional follow up time- 2 months
- Data analysis -1 month

### *5.3 Target population*

All pregnant HIV infected women presenting to the University Teaching Hospital.

### *5.4 Study population*

HIV infected pregnant women with CD4 counts of up to 350cells/$mm^3$ commenced on nevirapine based antiretroviral therapy in current pregnancy.

### *5.5 Selection of subjects*

#### 5.5.1 Inclusion criteria

- Pregnant women with documented HIV-positive status with CD4 counts less than or equal to 350 cells/$mm^3$ commenced on nevirapine based regimen in current pregnancy
- More than 18 years and able to give informed consent
- Willingness to attend antenatal and postnatal care at UTH

#### 5.5.2 Exclusion criteria

- Patients who are less than 18 years old or unable to give informed consent
- Patients who have been on cumulative 7 days or more of HAART

### *5.6 Study site*

The study was undertaken at the University Teaching Hospital (UTH) Department of Obstetrics and Gynaecology in the Adult Infectious Disease Center (AIDC).

### *5.7 Description of the study site*

UTH is the largest as well as highest referral hospital in Zambia. Although primarily a referral center, UTH also provides care to self-referred patients. It therefore has a wide spectrum of patients in terms of social-economic status and educational background.

Antenatal patients are seen in the UTH antenatal care clinic. After passing through the clerk's desk, patients receive different counseling sessions (conducted by midwives) before they are seen by the doctors. After the counseling sessions, opt out HIV testing is offered as part of the routine antenatal package.

Women who tested positive as well as those who already had a documented HIV positive status were routinely examined to asses clinical stage of the disease and bloods taken for CD4 counts and other baseline investigations ( FBC, LFTs, Creatinine, RPR, hepatitis B).The patients were then referred to the UTH ART-antenatal care clinic at the Adult Infectious Disease Centre (AIDC).

The study followed up women with CD4 counts less than or equal to 350 cells/$mm^3$ who were commenced on nevirapine-based antiretroviral therapy for about 12 to 14 weeks in order to evaluate their risk of nevirapine toxicity.

## 6 SAMPLING

Convenience sampling was used to enroll 146 patients into the study. All HIV-infected pregnant women with CD4 counts less than or equal to 350 cells/$mm^3$ commenced on nevirapine based antiretroviral therapy seen during the study period where enrolled if they consented and they fulfilled the eligibility criteria. Women with CD4 counts less than 250 cells/$mm^3$ commenced on nevirapine based regimen were allocated to group 1 arm and women with CD4 counts between 250 and 350 cells/$mm^3$ commenced on nevirapine based regimen in this pregnancy, to the group 2 arm.

### *6.1 Sample size calculation*

1) **Alternative hypothesis (one sided):** The incidence of nevirapine toxicity in HIV infected pregnant women who start HAART with CD4 counts between 250 and 350cells/$mm^3$ is higher than that of women who start with CD4 counts < 250cells/$mm^3$.
2) **P1** = from the literature, the incidence of nevirapine toxicity in HIV infected pregnant women who start HAART with CD4 counts less than 250cells/$mm^3$ is about **1% .**
3) **P2** = from the literature, the incidence of nevirapine toxicity in HIV-1 infected pregnant women who start HAART with CD4 counts greater than 250cells/$mm^3$ is about **16%.**
4) **Alpha** (one sided) = **0.05**.
5) At 80% power; **Beta** = 1 – 0.80 = **0.20**.

To detect a **15% difference** in the incidence of nevirapine toxicity in the two arms, the required sample size was 71 participants in each arm i.e. a total of 142 participants. This was adjusted to **146** patients at 10% loss to follow up per year.

## 7 STUDY PROCEDURE

HIV-infected pregnant women with CD4 counts less than or equal to 350cells/$mm^3$ seen at the study site were commenced on nevirapine based antiretroviral drug regimen and followed up for about 3 months in order to monitor the development of nevirapine toxicity. These patients were enrolled onto the study upon giving consent.

On the **first visit** all baseline data was collected including; documented HIV positive status, WHO clinical staging of the disease, CD4 count and baseline investigations including FBC, ALT, RPR, and Hepatitis B surface antigen test. Clinical staging and baseline investigations were done if these have not been done already.

Other routine antenatal investigations such as blood typing, RPR and urinalysis were also carried out if these hadn't been done. Mebendazole, haematinics for treatment and prevention of anemia, Cotrimoxazole and vitamins were given.

On her **second visit** at the ART clinic, drugs for HAART were dispensed. Adherence counseling as well as counseling on feeding options and safer sex was carried out.

The woman was given an appointment to return to the ART-antenatal clinic in 2 weeks to determine how she was doing.

On the **third visit,** the study assistant would look out for any rash as well as other side effects such as nausea, vomiting and anaemia. Hemoglobin was done and the nevirapine dose escalated.

The **subsequent visits** (3 visits) were after every 4 weeks. The study assistants continued to look out for any rash and took laboratory samples for analysis (AST, urinalysis) at each visit. Though plans were in place to manage patients with severe toxicity as in-patients, none of the patients had toxicity requiring in-patient care.

**Summary of visits**

| | |
|---|---|
| 1st visit | -Clinical evaluation<br>-Counseling (adherence, feeding options, safer sex). -Collected blood for CD4 count and baseline investigations (FBC, ALT, RPR, and Hepatitis B surface antigen test) |
| 2nd Visit (1 wk from last visit) | -Commenced NVP-based ART (any gestation)<br>-Counseling-side effects.<br>-Consent signed |
| 3rd Visit (2 weeks) | -Examined for rash.<br>-Collected blood for Hb.<br>-NVP dose escalated.<br>-Continued counseling. |

| | |
|---|---|
| 4th visit (4 weeks) | -Collected blood for ALT, Hb.<br>-Examined for rash.<br>-Counseling.<br>-Commenced Cotrimoxazole. |
| 5th visit (4 weeks) | -Examined for rash.<br>-Collected blood for ALT, Hb.<br>-Counseling. |
| 6thVisit (4 weeks) | -Examined for rash.<br>-Collected blood for ALT, Hb.<br>-Counseling. |

## 8 DATA COLLECTION TOOLS

Data was collected by the principle investigator and study assistants. A data clerk was responsible for data entry. All study personnel undertook a one week training of the study logistics prior to commencing the study.

For all participants, demographic details were obtained at the recruitment stage using an interviewer administered questionnaire. On the subsequent visits, the study assistant examined the participants for rash and looked out for signs of hepatic toxicity. A checklist was used.

### *8.1 Data management*

All study personnel had a one week training of the study logistics prior to starting the study. The data collected at various stages of the study was verified before being entered. A computerized data base was established. Double entry by two data clerks, range checks and consistency checks were carried out to reduce on errors.

## 9 DATA ANALYSIS AND STATISTICAL METHODS

**Independent variable; CD4** count

**Dependant variable (outcome variable);** Nevirapine toxicity (either cutaneous and/or Hepatotoxicity)

**Exposure;** Nevirapine-based HAART regimen in pregnancy

**Nevirapine toxicity** defined as:

1. Rash- at least grade 2 rash with or without mucous membrane involvement.
2. Hepatic toxicity – at least grade 2 rise in aspartate amino transferase or alanine amino transferase

The grading of toxicity was as per WHO toxicity estimates (2006 WHO ART guidelines)

The incidence of nevirapine toxicity in both study arms was obtained and compared. The proportions were compared using the Z test. Multivariable analysis was done for different variables in secondary analysis. SAS statistical package was used in the analysis.

## 10 ETHICAL CONSIDERATIONS

Ethical approval to carry out the study was obtained from the University of Zambia Research Ethics Committee (UNZAREC). Patients were enrolled on voluntary basis after completing a detailed consent form.

Privacy and confidentiality were maintained. Each patient was given an identification number different from her ZEPRS and antenatal card numbers. Patient's records were only accessible to study personal. National guidelines on HIV testing and counseling were adhered to. The information provided by study participants including laboratory tests was treated as confidential. The study did not cause any undue delays and patients did not incur any additional costs as a result of the study.

All HIV positive pregnant women who refused to give consent for the study continued antenatal care and were managed like any other patients, without any limitations to medical treatment.

### *10.1 Benefits of the study*

The study has added to the body of knowledge, information regarding nevirapine-based antiretroviral therapy in pregnancy. Policy makers have been provided with local, research based information that can influence policy with a view to scaling up best practices for PMTCT in resource constrained areas. Participants had an opportunity to receive a comprehensive antenatal and PMTCT package with clinical and laboratory follow-up.

## 11 RESULTS

Data collection took place at AIDC in UTH. Data collection was complete by 5th February 2010.

### *11.1 Social demographic data*

A total of 148 women were analyzed. The age distribution is shown below. The average was 30.8, median 31 and the range 18 to 43 years. Four women were 40 or more years old.

| Analysis Variable : Age | | | | | | |
|---|---|---|---|---|---|---|
| | N | Mean | Std Dev | Median | Minimum | Maximum |
| | 148 | 30.8 | 5.1 | 31 | 18 | 43 |

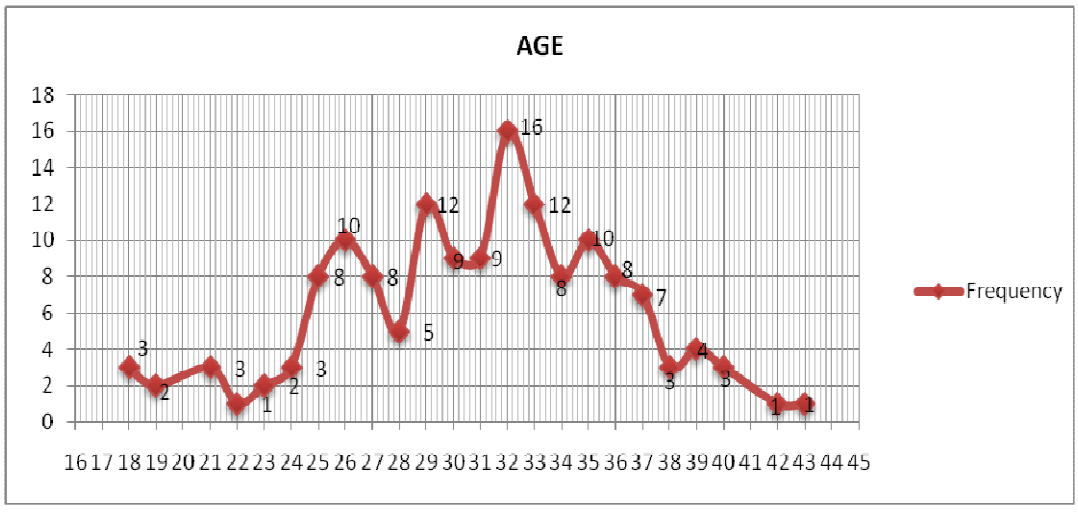
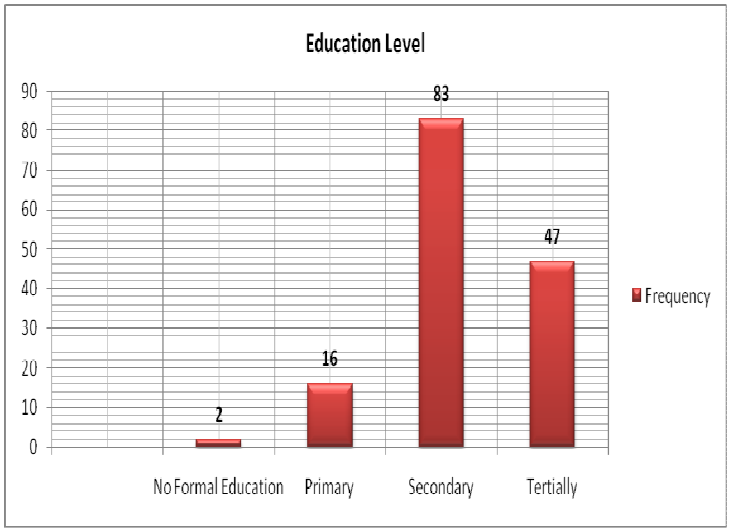



Figure 1: Age distribution of women

**Education background**

Total number recorded = 148



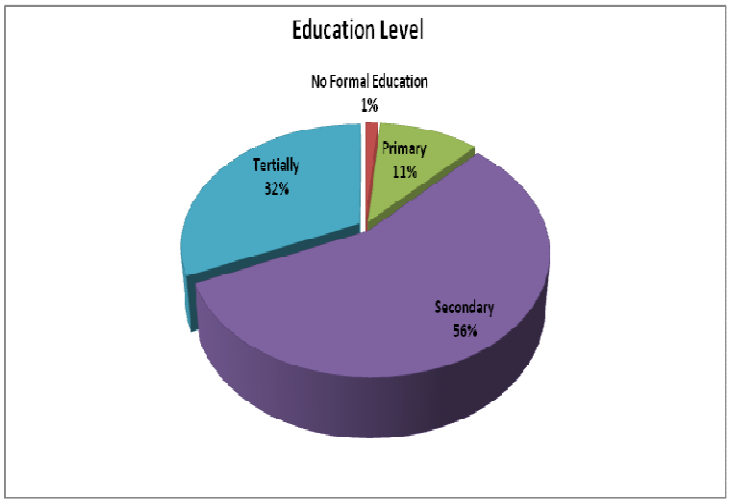




Figure 2: Educational background of women enrolled

Majority, 83 of 148 (56%) had up to secondary education. Only 47 of 148 (32%) women had tertially education. 11% had primary education while 1% had no formal education.

**Occupation**

Total recorded = 148

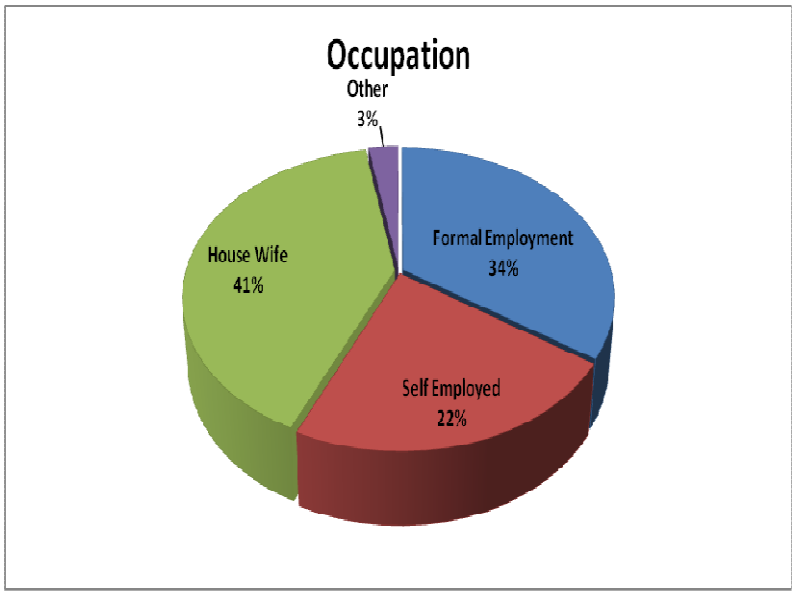
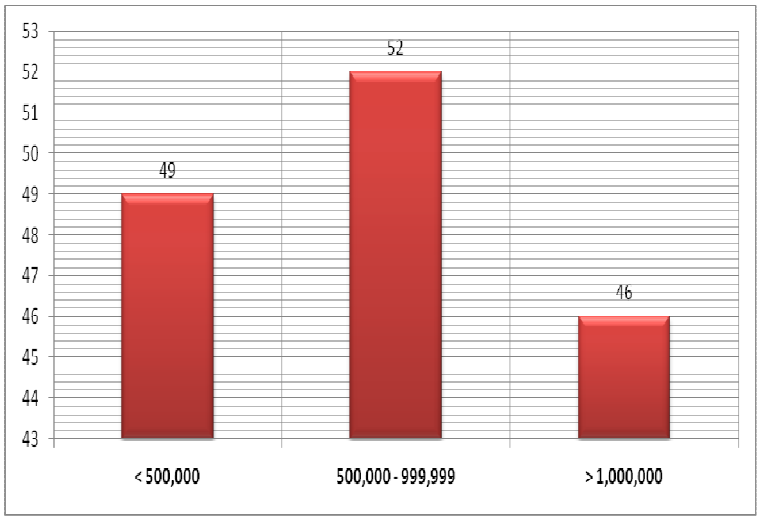




Figure 3: Occupation of women enrolled

The women in the `other` category were unmarried woman in school or unmarried dependants not involved in any income generating activity.

**Total household income**

Total recorded = 148



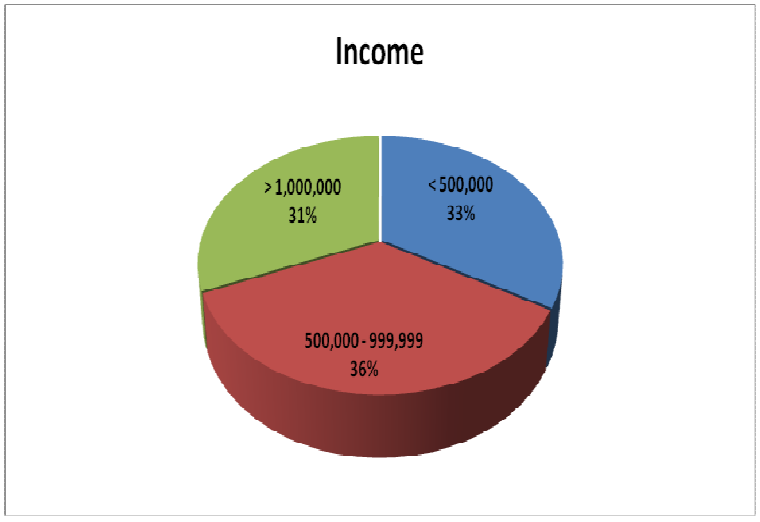
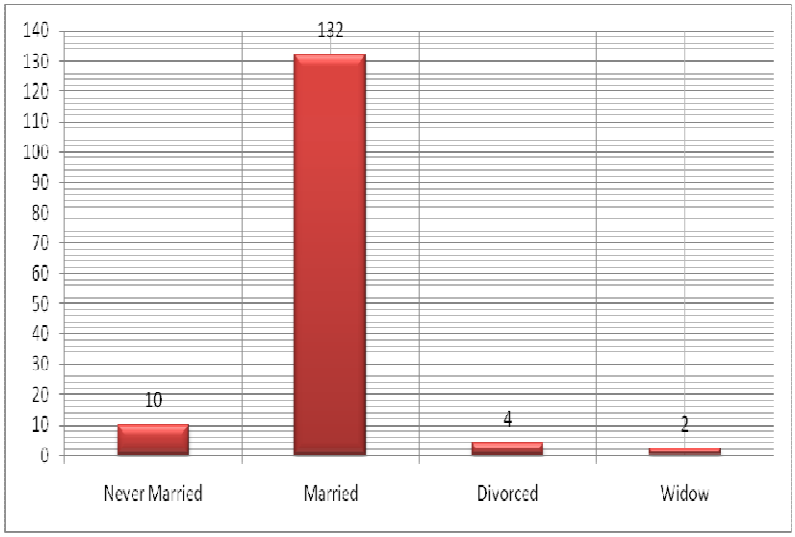




Figure 4: Total household income for women in the study

49 (33%) out of 148 women came from households whose income was less than Five hundred thousand kwacha. 46 (31% from households with income more than one million kwacha.

**Marital status**

Total recorded = 148



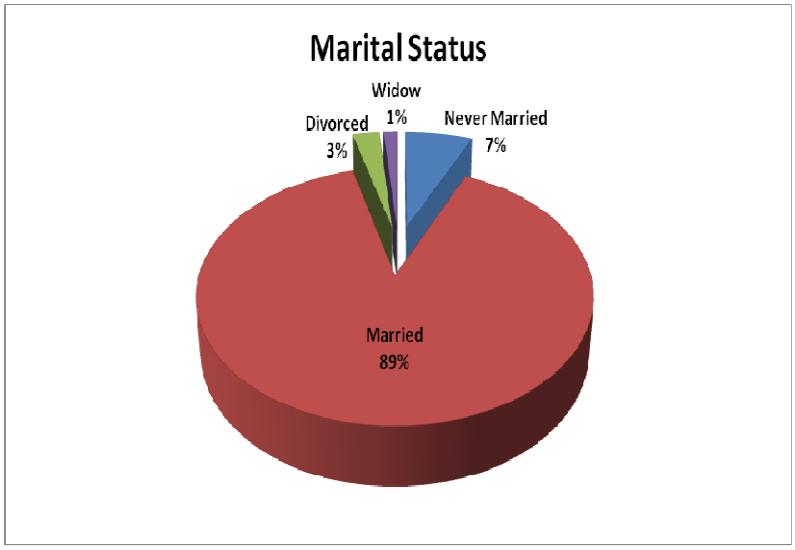
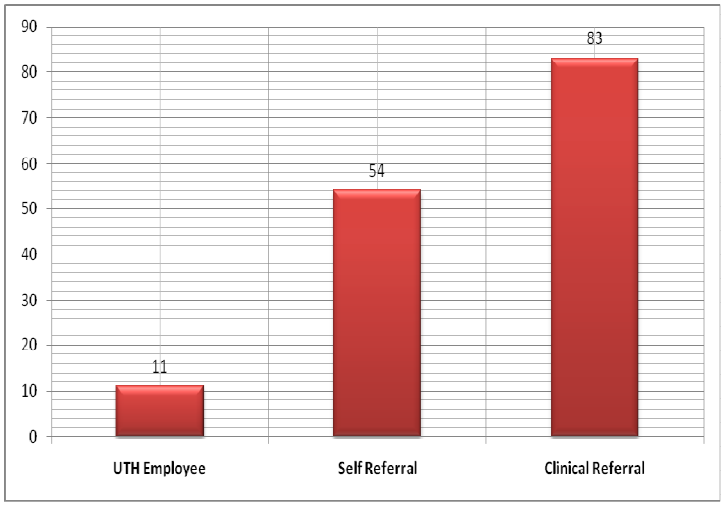




Figure 5: Marital status of women

132 (89%) of 148 patients were married, 16 (11%) where single parents.

**Patients Referral to the AIDC clinic**

Total recorded = 148



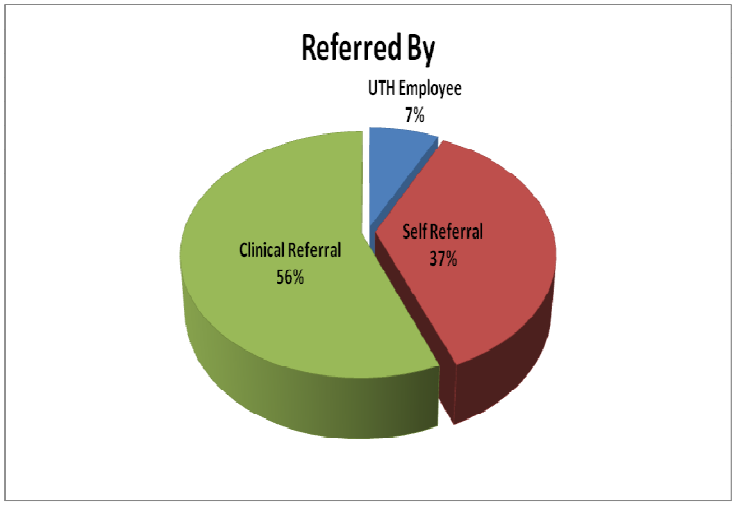
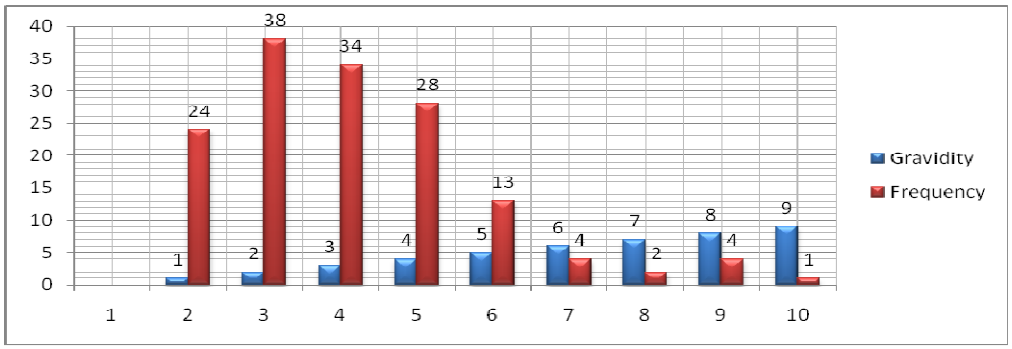




Figure 6: Patients` Referral to the AIDC clinic

83 (56%) of 148 patients were referred to UTH for further management. 54 (37%) of 148 patients were paying patients who are self referred. 11 (7%) of the patients were either UTH employees or spouses of UTH employees

**Gravidity**

| Analysis Variable : Gravidity | | | | | |
|---|---|---|---|---|---|
| N | Mean | Std Dev | Median | Minimum | Maximum |
| 148 | 3.1 | 1.7 | 3 | 1 | 9 |




**Parity**

| Analysis Variable : Parity | | | | | |
|---|---|---|---|---|---|
| N | Mean | Std Dev | Median | Minimum | Maximum |
| 148 | 1.8 | 1.5 | 2 | 0 | 8 |

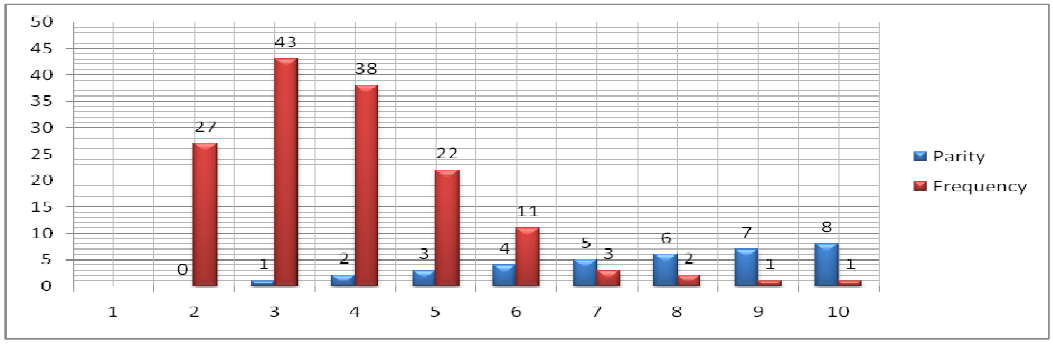



Figure 7: Gravidity and parity

The highest gravidity was 9 with an average of 3.1 pregnancies while the highest parity was 8 with an average of 1.8 deliveries.

**Number of children alive**

Total number of children = 146

| Analysis Variable : No of children alive | | | | | |
|---|---|---|---|---|---|
| N | Mean | Std Dev | Median | Minimum | Maximum |
| 146 | 1.7 | 1.2 | 2 | 0 | 6 |

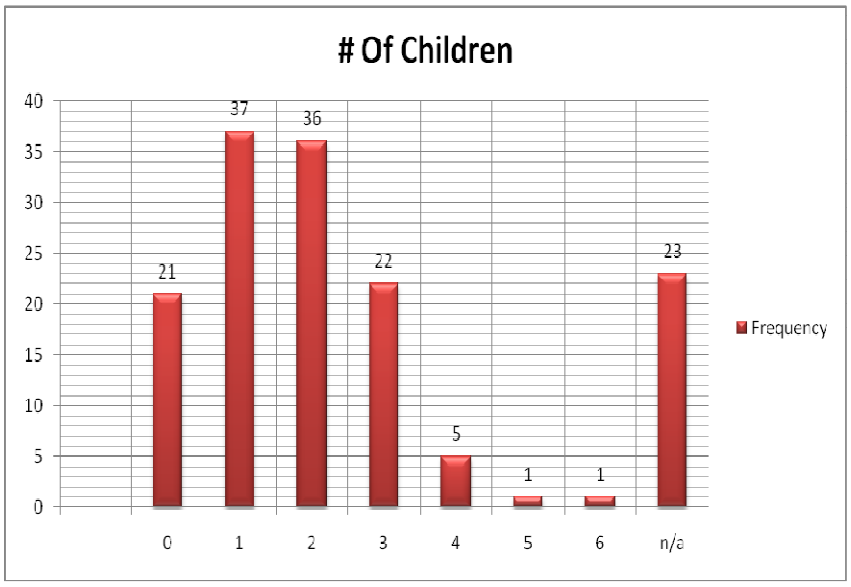



Figure 8: Number of children alive

The `not applicable` category where women having their first pregnancy while the 0 number of children are women who have been pregnant before but have no living children

**Gestation at enrollment onto HAART**

Total number 148

| Analysis Variable : Gestation In Weeks | | | | | |
|---|---|---|---|---|---|
| N | Mean | Std Dev | Median | Minimum | Maximum |
| 148 | 28.9 | 7.1 | 29.5 | 9 | 41 |

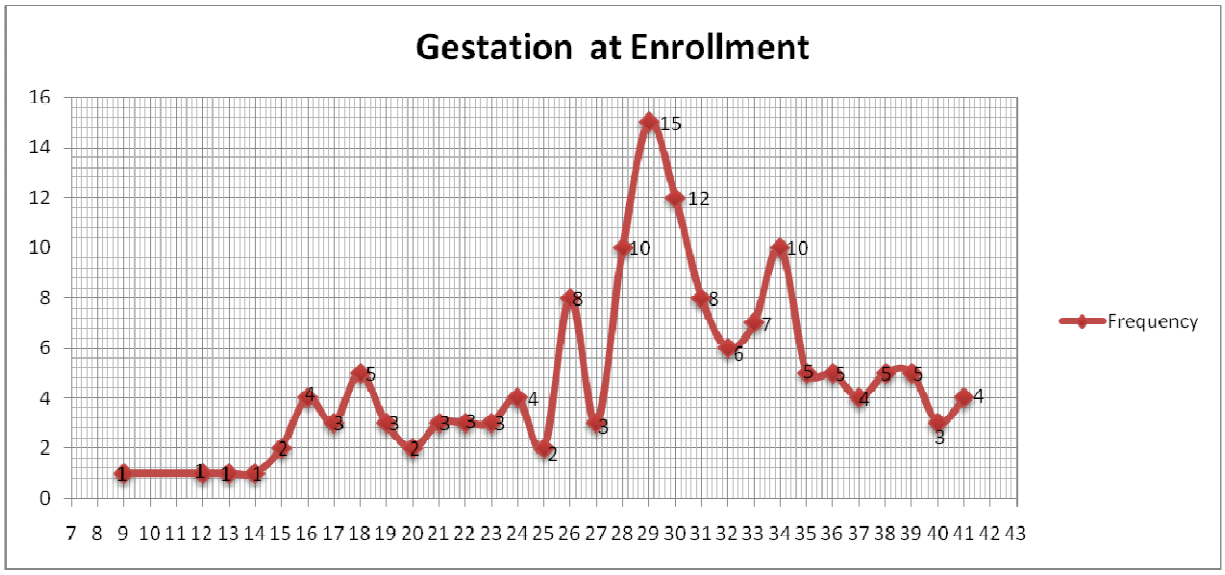
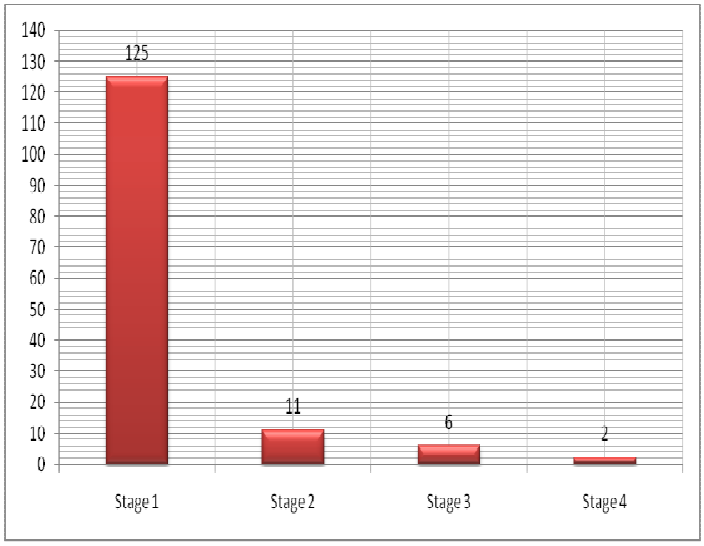



Figure 9: Gestation at enrollment into HAART

The mean gestation at enrollment was 28.9 weeks, the median was 29.5 weeks. 4 patients were enrolled onto HAART at 41 weeks gestation.

**W.H.O clinical stage of disease at enrollment**

Total number 144



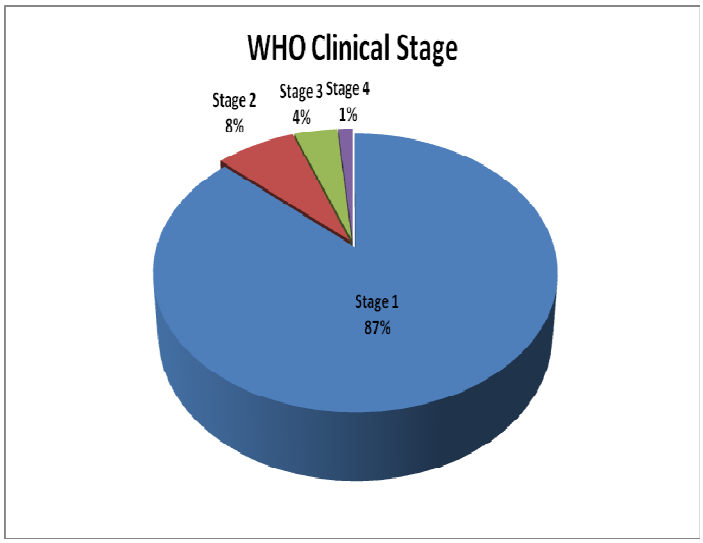




Figure 10: W.H.O clinical stage of disease at enrollment

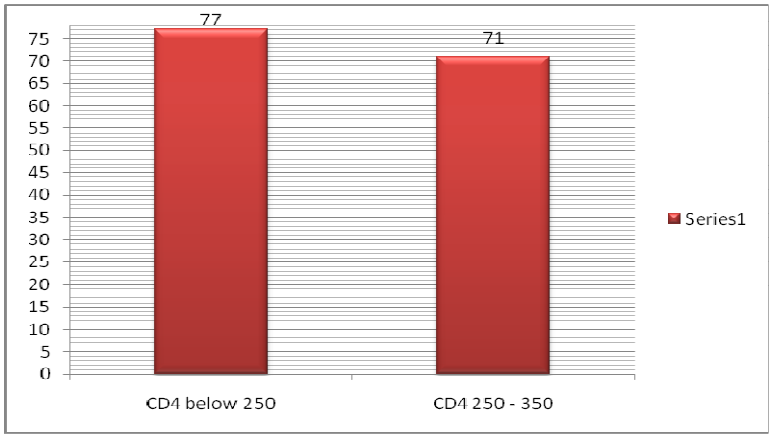
125 (87%) of 144 women had W.H.O clinical stage 1 disease at enrollment into Highly Antiretroviral Therapy (HAART)

**CD4 counts at enrollment**

Total number of patients: 148.

| Analysis Variable : InitialCD4 | | | | | |
|---|---|---|---|---|---|
| N | Mean | Std Dev | Median | Minimum | Maximum |
| 148 | 223.4 | 82.2 | 240.5 | 39 | 350 |

Proportion of CD4 counts below 250, to that of CD4 counts 250 to 350



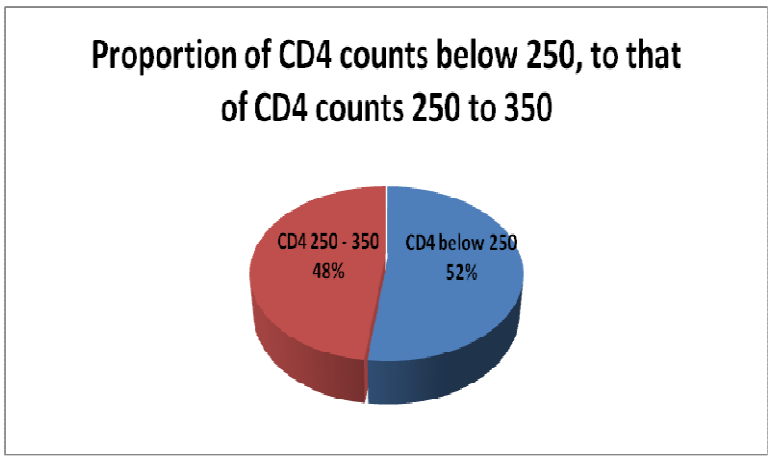
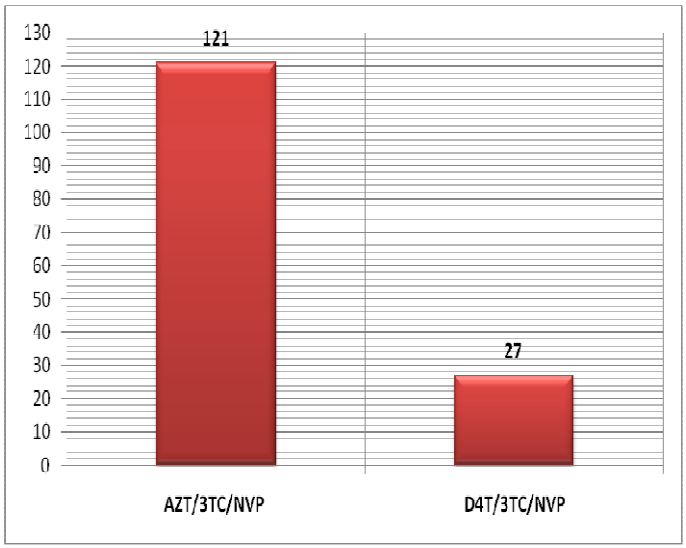




Figure 11: Proportion of women with CD4 counts below 250, to that of women with CD4 counts 250 to 350cell/mm$^3$

**Proportion of patients on AZT/3TC/NVP to those on D4T/3TC/NVP**

Total number of patients 148



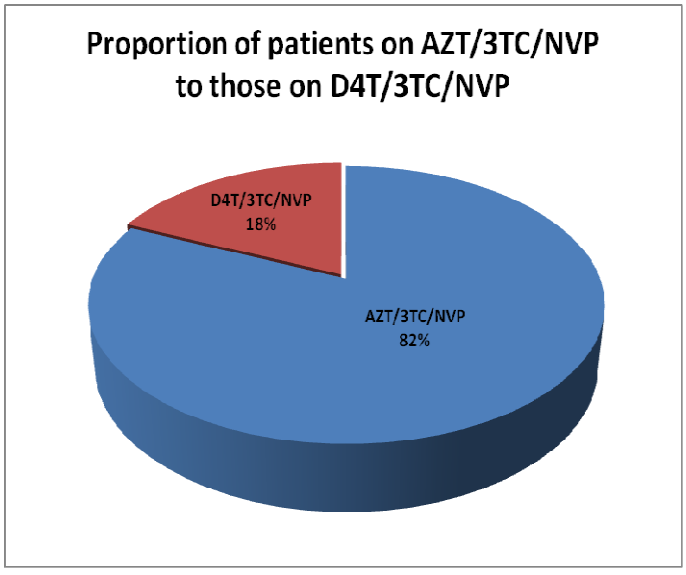
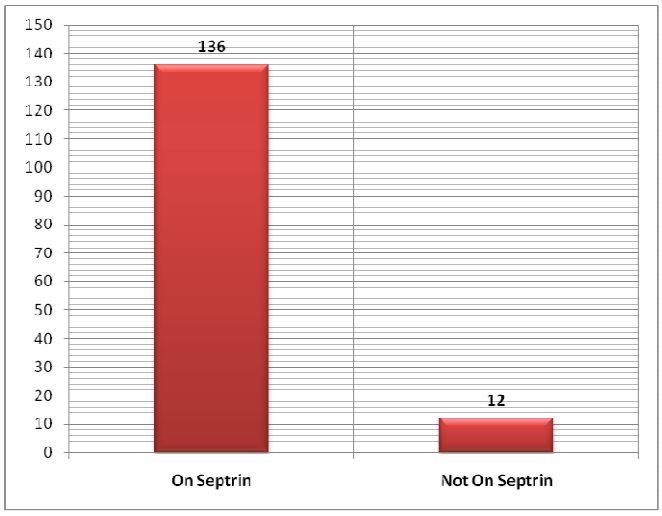




Figure 11: Proportion of patients on AZT/3TC/NVP to those ON D4T/3TC/NVP

**Proportion of patients on Septrin**

Total number of patients 148



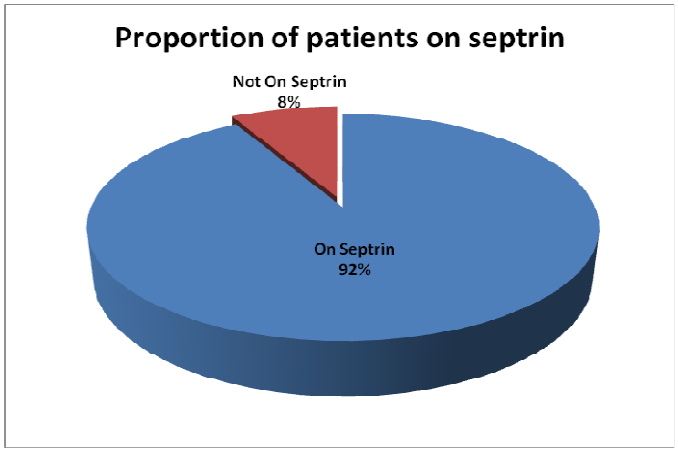
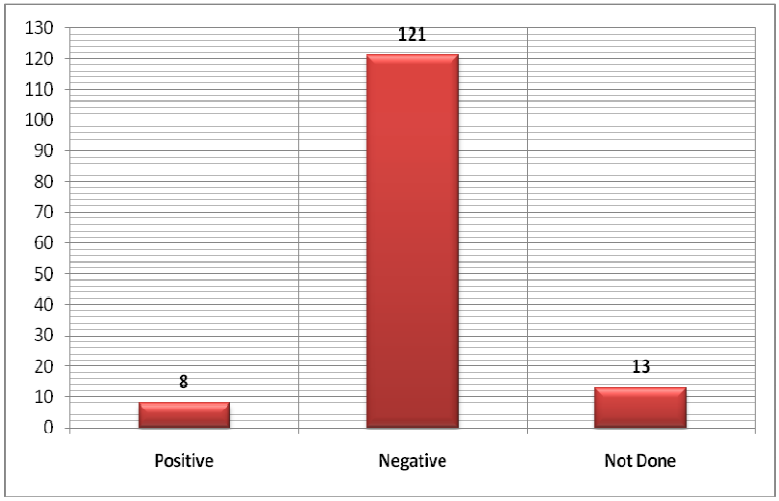




Figure 12: Proportion of patients on Septrin

136 (92%) of 148 patients had been commenced on Septrin

### *11.2 Prevalence of Hepatitis B co-infection*

Total number of patients Recorded 142



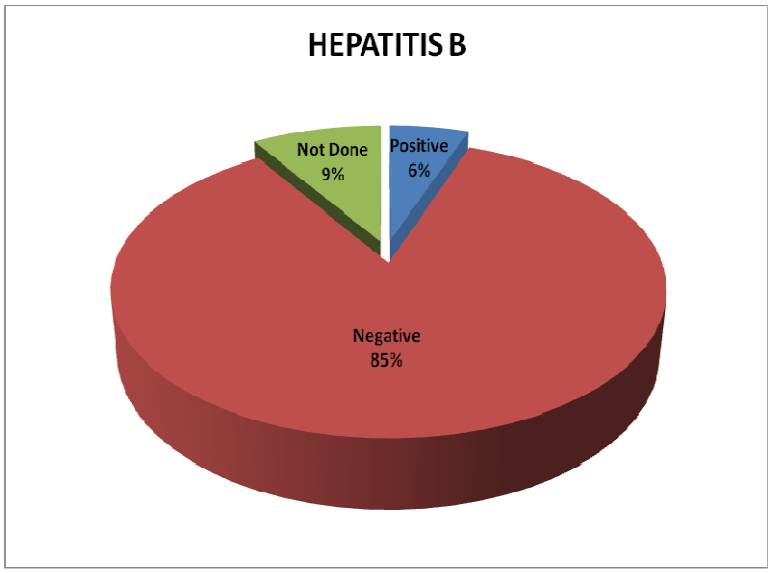
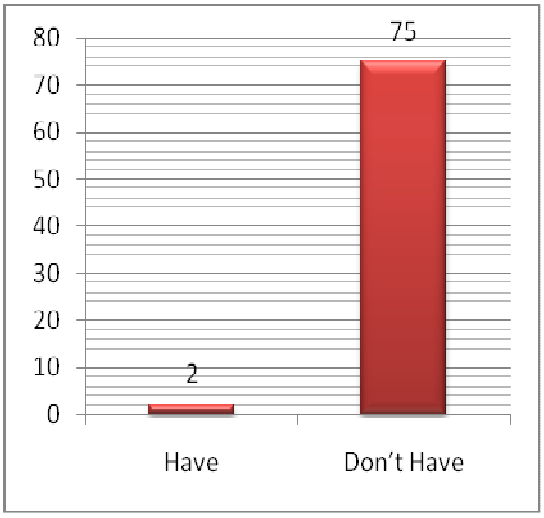



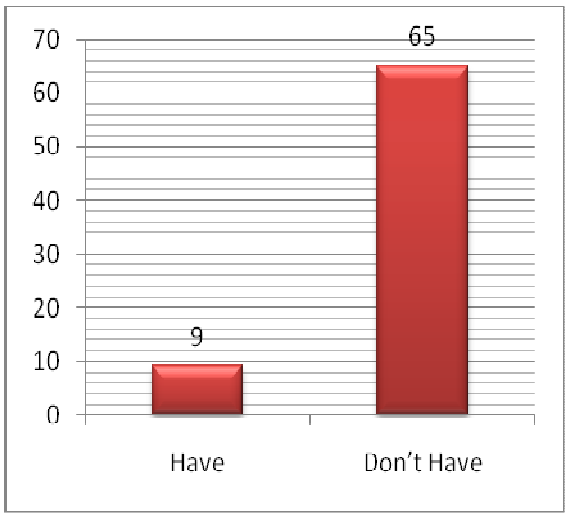

Figure13: Patients with hepatitis B co-infection

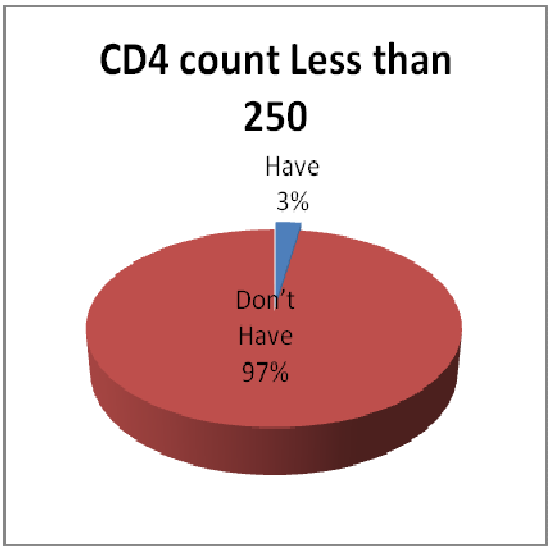
8 (6%) of the 142 patients had hepatitis B co-infection

### *11.3 Incidence of hepatic toxicity (deranged aspartate aminotransferase levels )*

CD4 count Less than 250 CD4 count 250 to 350



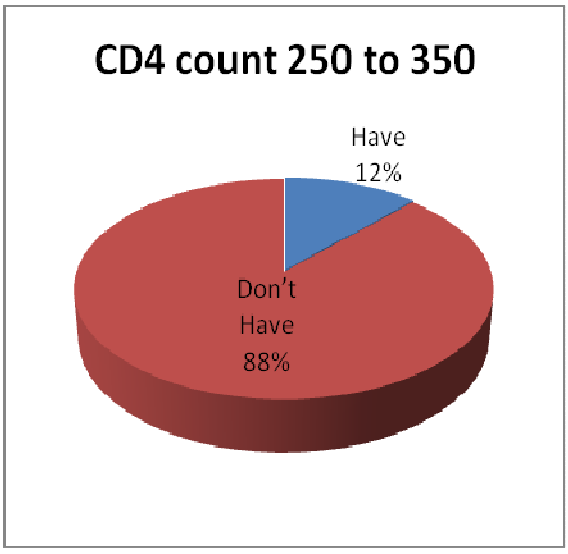
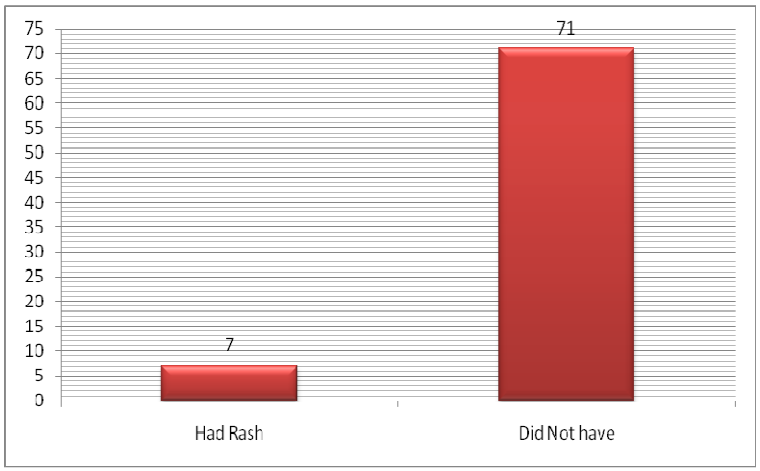




Figure 14: Patients who developed NVP rash.

2 (3%) of 77 patients with CD4 count less than 250cells/mm$^3$ had deranged liver function while 9 (12%) of 74 patients with CD4 counts between 250 to 350cells/mm$^3$ had deranged liver function.

### *11.4 Incidence of cutaneous toxicity (Patients with nevirapine rash)*

NVP rash for CD4 Count 250 to 350cell/mm$^3$



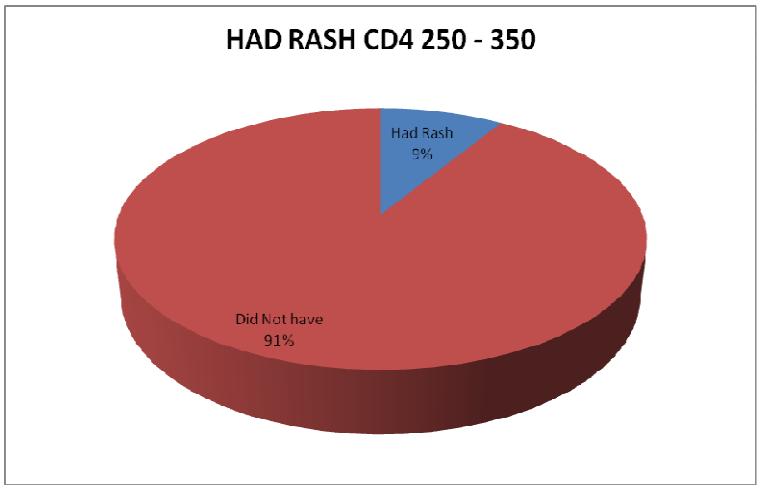




Figure 15: Patients with nevirapine rash

No patient with CD4 count less than 250 had skin rash.

7 (9%) of the 71 patients with CD4 counts of 250 to 350 had generalized maculopapular rash after being commenced on Nevirapine based regimen.

# 12 DISCUSSION

The aim of the study was to evaluate the incidence of nevirapine toxicity in pregnant HIV-infected women commenced on nevirapine based regimen in the current pregnancy with CD4 counts less than or equal to 350 cells/$mm^3$ at the University Teaching Hospital, Lusaka, Zambia. It also aimed to compare the incidence of nevirapine toxicity in women with CD4 counts less than 250 cells/$mm^3$ to that of women with CD4 counts of 250 to 350 cells/$mm^3$ as well as to evaluate the prevalence of hepatitis B co-infection in the study population.

The incidence of hepatic toxicity was 8%.That of cutaneous toxicity was about 7%. The combined incidence was 13.5%. The prevalence of hepatitis B co infection was 6%.

The findings in this study may be used in accessing the safety of nevirapine-based regimen in HIV positive pregnant women. As Ministry of Health rolls out the PMTCT program, more pregnant women are likely to commence HAART. More women with CD4 counts less than 350 cells/$mm^3$ will be exposed to nevirapine based highly active antiretroviral therapy regimen as this is the recommended first line regimen.

## *12.1 Social demographic data*

The age distribution of women in the study ranged from 18 to 43 years with an average of 30.8 years. Fifty six percent (56%) had only up to secondary school education while 1% had no formal education. Thirty four (34%) of women were in formal employment and 22% where self employed. The low educational levels and the low employment levels

contribute to the low income levels with about 33% of women coming from households with less than five hundred thousand kwacha total income per month.

Eighty nine percent (89%) of the women were married. The figure represents women whose partners are either already infected or are at very high risk of being infected. The huge number of married women reflects the need for PMTCT programs to rigorously incorporate other aspects of ART care such as partner notification. While antenatal care is an opportunity for client entry into other services such as primary health care, family planning, screening for cervical cancer etc, it is also an opportunity for entry of these women`s husbands and children into ART care and ART-related services such as counseling and social support.

Fifty six percent (56%) of women were seen at the AIDC clinic by reason of being referred to the University Teaching Hospital for complicated care while 37% where fee paying patients in search of better care.

The mean gravidity was 3.1 with a maximum of 9 pregnancies while the mean parity was 1.8 with a maximum of 8. The average number of children was 1.5. The progressive decrease from gravidity, parity to the number of children alive may be explained by the relatively higher incidence of pregnancy loses (miscarriages and still births) and pediatric deaths associated with the HIV infection.

Figure 9 shows the average gestation at enrollment was about 29 weeks, with 4 women enrolling at 41 weeks. Being referred may be a contributing factor to late enrollment as 56% of the women were referred from local clinics for complicated care. However, pregnant women and their partners need to be sensitized regarding the benefits of starting ART sooner in order to maximize outcome for both the mother and the baby.

Seventy seven (52%) women had CD4 counts less than 250 cells/$mm^3$ while seventy one (48%) women had CD4 counts between 250 and 350 cells/$mm^3$ at enrollment. There was poor correlation between the WHO clinical stage and the CD4 count in the 2 cohorts as 87% of women enrolled had WHO clinical stage 1 disease. Eighty two percent (82%) of women were commenced on AZT/3TC/NVP. The 27 patients (18%) commenced on D4T/3TC/NVP had hemoglobin less than 10g/dl when starting HAART. Ninety two percent (92%) of patients took cotrimoxazole prophylaxis as per the 2007 Ministry of Health, Zambia ART guidelines. Four (4) of the 6 patients who did not take cotrimoxazol had reported prior allergy to the drug.

### *12.2 Prevalence of Hepatitis B co-infection*

8 (6%) of 142 patients had hepatitis B co-infection. These were clinically stable patients with normal baseline aspartate alanine transferase before commencing HAART. 2 of the 8 patients with hepatitis B co-infection had deranged liver function tests grade 1 and grade 2 respectively after commencing nevirapine based HAART regimen.

### *12.3 Incidence of hepatic toxicity (deranged aspartate aminotransferase levels)*

2 (3%) of 77 patients with CD4 count less than 250cells/$mm^3$ had deranged aspartate aminotransferase. Both patients had grade 1 elevation of the enzyme.

9 (12%) of 74 patients with CD4 counts between 250 to 350cells/$mm^3$ had altered aspartate aminotransferase. 3 of the nine patients had grade 1 elevation in ALT while 6 (8%) patients had grade 2 or more elevations in ALT.

### *12.4 Incidence of nevirapine rash*

No patient with CD4 count less than 250cells/mm3 developed rash after being commenced on nevirapine based HAART regimen.

7 (9%) of the 71 patients with CD4 counts of 250 to 350 had generalized maculopapular rash after being commenced on Nevirapine based regimen. 1 Patient had grade 1 rash. She was managed conservatively and continued on the same regimen. The other 6 (7%) had either grade 2 or 3 rash. None of the patients developed grade 4 rash and none were admitted. Nevirapine was promptly discontinued if rash is grade 2 or more. Steroids and antihistamines were given and all patients recovered well.

### *12.5 Incidence of Nevirapine toxicity (combined hepatic and cutaneous toxicity)*

The incidence of nevirapine toxicity (defined as either 1and/or 2):

1. Rash- at least grade 2 rash with or without mucous membrane involvement.
2. Hepatic toxicity – at least grade 2 rise in Aspartate Amino Transferase or Alanine Amino Transferase

Nevirapine toxicity in the CD4 count arm less than 250 was 0%.

In the CD4 count arm 250 to 350 cells/$mm^3$; 6 of 74 patients had grade 2 or more elevation in ALT and 6 of 71 patients had grade 2 or more maculopapular rash. 2 of these patients had both elevation in ALT and rash. Therefore the incidence of nevirapine toxicity is 13.5% *(p*= 0.005) in this study population.

## 13 CONCLUSIONS

The results show that, the incidence of nevirapine toxicity in this study was at 0% in women with CD4 counts below 250cells/$mm^3$

In women with CD4 counts between 250 to 350cell/$mm^3$, the incidence of nevirapine toxicity was 13.5%. Though the study was powered to detect a 15% difference, the results of the study show that women with CD4 counts of 250 to 350cells/$mm^3$ were at substantial risk of nevirapine toxicity when commenced on nevirapine based HAART regimen in pregnancy. Prompt discontinuation of the drug and supportive therapy may have contributed to prevention of further progression to grade four toxicity.

Hepatitis B co-infection does not seem to contribute to the incidence of nevirapine toxicity in this study population

## 14 STUDY LIMITATIONS

Loss to follow up was expected as patients were followed up for a period of about 3 months.

## 15 STRENGTHS OF THE STUDY

A Longitudinal observational study is a reliable study design to detect and compare incidence of Nevirapine toxicity in the 2 study arms. The study was carried out within existing structures-The University Teaching Hospital and the Adult infectious Disease Centre (AIDC) with technical support from both.

The results of the study are relevant to the population being studied and can be generalized to all HIV infected pregnant women with CD4 counts less than 350 cells/$mm^3$ who are commenced on Nevirapine based regimen in the current pregnancy.

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## ANNEXES

### Annex 1: Budget for Nevirapine toxicity in HIV-1 infected pregnant women study

| Budget Item | Description | Unit-Cost (USD) | Quantity | Total (USD) |
|---|---|---|---|---|
| ***Personal*** | | | | |
| | 1 obstetrician | | | |
| | 1 Statistician | | | |
| | 2 Data Clerks | | | |
| | 2 Nurses | | | |
| | 2 Counsellors | | | |
| ***Investigations*** | | | | |
| | Full Blood Count (FBC) | 10 /One test | 200X1 | 2000=00 |
| | CD4 count | 15 /One test | 200X1 | 3000=00 |
| | ALT<br>AST | 10/One test<br>10/one test | 200X4<br>200X4 | 8000=00<br>8000=00 |
| | Creatinine | 8/One test | 200X1 | 1600=00 |
| | RPR | 5/One test | 200X2 | 2000=00 |
| | Blood Typing | 5/One test | 200X1 | 1000=00 |
| | Hb | 5/One test | 200X3 | 600=00 |
| | Hepatitis B sAg | 10/one test | 200X1 | 2000=00 |
| | Urinalysis | 5/One test | 200X7 | 7000=00 |
| ***Drugs*** | | | | |
| | HAART | 30/ month | 200X5 | 30000=00 |
| | Cotrimoxazole | 5/ month | 200X5 | 5000=00 |
| | Mebendazole | 5/ month | 200X5 | 5000=00 |
| | Iron, Folic Acid, MVT | 5/ month | 200X5 | 5000=00 |
| ***Equipment*** | | | | |
| | Blood pressure machines | 200/ each | 200X2 | 400=00 |
| | Computers | 1000/each | 1000X2 | 2000=00 |
| | Printer | 500/each | 500X1 | 500=00 |
| | Phone | 200/each | 200X2 | 400=00 |

| Budget Item | Description | Unit-Cost (USD) | Quantity | Total (USD) |
|---|---|---|---|---|
| ***Others*** | | | | |
| | Stationary | 10/each | 2X12 | 240=00 |
| | Transport, talk time | | | 1000=00 |
| **TOTAL** | | | | **84,740** |

**Annex 2: Budget Justification**

| **Budget item** | **Source of funding** |
|---|---|
| Personal-employed by MOH/CIDRZ | MOH/CIDRZ |
| Investigations- all investigations are routine investigations done (or suppose to be done) on all pregnant HIV positive patients | MOH/CIDRZ |
| Drugs- all drugs are routine drugs given (or suppose to be given) to HIV positive patients attending antenatal clinic at UTH | MOH/CIDRZ |
| Equipment-already present at AIDC | CIDRZ |
| Others | CIDRZ/self |

Note: *MOH- Ministry of Health, Zambia. CIDRZ- Centre for Infectious Disease Research, Zambia. The amounts given for investigations and drugs are the cost of items in private hospital settings*

**Annex 3: WHO cutaneous toxicity estimates**

| Toxicity | Clinical description | Recommendation |
|---|---|---|
| **Grade 1** | Erythema | Continue nevirapine with careful and repeated observation and follow up. Provide symptomatic treatment such as antihistamines |
| **Grade 2** | Diffuse maculopapular rash, dry desquamation | Continue nevirapine with careful and repeated observation and follow up if possible. Provide symptomatic treatment. If no improvement, consider substitution. |
| **Grade 3** | Vesiculation, mucosal ulceration | Substitute nevirapine |
| **Grade 4** | Exfoliative dermatitis, Steven-Johnson Syndrome or erythema multiforme, moist desquamation | Discontinue all the ARV drugs, manage the medical event until patient is stable and toxicity has resolved |

*Adapted from WHO. Antiretroviral Drugs for Treating Pregnant Women and preventing HIV infection in infants in resource-limited settings; Towards Universal Access. Geneva; 2006*

**Annex 4: WHO hepatotoxicity estimates**

| | LFT range U/L | What to do: |
|---|---|---|
| **Normal** | | Continue medications unless pt symptomatic such as rash in patient taking NVP |
| | | |
| **Grade 1-2** | Grade I ALT; 40-78<br>AST; 40-80 | If pt on NVP/EFV with no clinical symptoms, then can continue medication with close follow up and repeat LFT testing |
| | Grade II ALT; 81-155<br>AST;83-160 | |
| **Grade 3-4** | Grade III ALT; 158-310<br>AST;163-320 | Stop all ART. Can restart once LFTs go down to grade 1. Do not restart NVP. |
| | Grade IV ALT; >310<br>AST; >320 | |

*Adapted from WHO. Antiretroviral Drugs for Treating Pregnant Women and preventing HIV infection in infants in resource-limited settings; Towards Universal Access. Geneva; 2006*

**Annex 5: Zambian Criteria for PMTCT intervention**

| | **Provide HAART** | **Provide short course ARVs** |
|---|---|---|
| **CD4** | <350/mm$^3$ | >350/mm$^3$ |
| **Clinical criteria only(CD4 not available)** | Stage 3 or 4 (any CD4[a]) | Stage 1 or 2 |
| **Gestational age** | <36 weeks if eligible for HAART or clinical criteria [b] | >32 weeks if ineligible for HAART [c, d] |

[a] if CD4>350 then initiate ART after 14 weeks with EFV plus 2 NRTIs, Pregnant women less than 14 weeks gestation should be given appropriate ARVs and monitored closely in the first trimester

[b] If the pregnant woman does not initiate HAART before 36 weeks then she should be given the short course of ARV prophylaxis (see also foot notes c and d)

[c] Check Hb, Should be above 8g/dl. Correct anaemia before giving short course ARV prophylaxis

[d] If the pregnant woman receives less than 4 weeks of AZT the baby should receive single dose of NVP + 4 weeks of AZT

*Adapted from Zambian MOH 2007 ART protocols*

## QUESTIONAIRE

Mark the appropriate response. Do not write respondent`s name on the Questionnaire

**Serial number** _____

| | | | |
|---|---|---|---|
| Q1 | Age | | |
| Q2 | Gravidity<br>*(Total number of pregnancies)* | | |
| Q3 | Parity<br>*Total number of deliveries after 28weeks gestation)* | | |
| Q4 | Number of Children alive | | |
| Q5 | Gestation<br>*From LNMP or scan* | | |
| Q6 | Where do you stay? | | |
| Q7 | Educational level | | |
| | No formal education | 1 | |
| | Primary | 2 | |
| | Secondary | 3 | |
| | Tertially | 4 | |
| Q8 | Marital status | | |
| | Single | 1 | |
| | Married | 2 | |
| | Separated | 3 | |
| | Divorced | 4 | |
| Q9 | Occupation | | |
| | Full time employee | 1 | |
| | part time employee | 2 | |
| | Business | 3 | |
| | Not employed | 4 | |
| | Student / pupil | **5** | |
| Q10 | How much do you earn per month? | | |
| Q11 | Referred by | | |
| | Self | **1** | |
| | Public clinic | **2** | |
| | Private clinic/hospital | **3** | |
| Q12 | Reason for referral | | |

## CHECKLIST

| | | | | | | |
|---|---|---|---|---|---|---|
| **Checklist for first visit** | | **Serial number----------** | | | | |
| Q1 | WHO clinical stage of disease | 1 | | 3 | | |
| | | 2 | | 4 | | |
| Q2 | CD4 count | | | | | |
| | <250cell/mm3 | | | 1 | | |
| | 250-350cell/mm3 | | | 2 | | |
| Q3 | Hepatitis B | | | | | |
| | Positive | | | 1 | | |
| | Negative | | | 2 | | |
| Q4 | Hemoglobin (Hb) | | | | | |
| Q5 | Aspartate amino transferase (AST) | | | 1 | | |
| | Alanine amino transferase (ALT) | | | 2 | | |
| Q6 | Creatinine | | | | | |
| Q7 | Rash | | | | | |
| | present | | | 1 | | |
| | Absent | | | 2 | | |
| Q8 | If rash is present-Grade? | 1 | | 3 | | |
| | | 2 | | 4 | | |
| **Check-list for subsequent visits** | | | | | | |
| **Serial number_____** | | | | | | |
| Q1 | Hemoglobin (Hb) | | | | | |
| Q2 | Aspartate amino transferase (AST) | | | | | |
| | Alanine amino transferase (ALT) | | | | | |
| Q3 | Rash | | | | | |
| | present | | | 1 | | |
| | Absent | | | 2 | | |
| Q4 | If rash is present-Grade? | 1 | | 3 | | |
| | | 2 | | 4 | | |

## CONSENT FORM

**Introduction**

Because you are pregnant and are infected with the human immunodeficiency virus (HIV, the virus that causes AIDS) and will be taking antiretroviral drugs (ARVs) for your health and also to prevent the HIV virus from infecting your baby, you are being asked to participate in this research project. You are being asked to take part in a research that will help collect information regarding your health while you are on these drugs. The information, such as side effects of the drugs, how well certain organs in your body are functioning will help answer questions about using these drugs in pregnancy.

**Procedures**

This study is designed to collect information about your health. As part of any routine antenatal care, you are asked questions related to your health, you are examined and blood samples are drawn. This will not change and no additional information or laboratory samples will be taken other than what is normally done or expected. Your care in the clinic will be both antenatal and PMTCT (Prevention of Mother to Child Transmission of HIV.) If you give permission to participate in this study, the investigators will collect and merge information from your clinic records if required, with the additional information you provide in the clinic.

**Risks, inconveniences, and discomforts**

Inconveniences include answering questions, typically for about 20 minutes (depending on how many questions we need to ask at a particular visit), during a regular visit to the clinic. The questions are about your general information, your health and the medicine(s) you are taking. You will be required to answer the questions as they are being asked. Someone will be available to help you if you need assistance.

You will be examined and blood drawing will be performed in the same standard way. No additional examinations or blood draws are required. The risks of having blood drawn are: bleeding or bruising at the needle entry site, hematoma (swelling in the tissue around the needle entry site) and rarely, obstruction of the blood vessel.

If you experience any form of stress from answering the questionnaires, clinical examination, or blood draw. You will be able to stop the process at any time. You could choose to continue later, to not continue, or to withdraw from the study completely at no penalty to you.

**Benefits**

The information collected in this study will be used to learn about the side effects associated with HIV drugs in pregnancy. While answering questions about the way you feel and how you are doing may provide some insights for you personally and for your caregivers into your health concerns, you may not otherwise benefit directly from this study. However, it is possible that combining the information from your health care experiences with that of other patients in this clinic, the investigators will be able to make improvements in the way that health care is provided to patients with HIV infection who are pregnant in the future, both at this clinic and elsewhere.

**Withdrawal without prejudice**

You are free to withdraw your consent and to discontinue participation in this project at any time without prejudice against future medical care at this institution.

**Alternatives to participation**

If you choose not to participate in this study, you may continue the medical care that you are currently receiving.

**Costs to you**
There will be no costs to you that are directly related to this study. You will continue to receive the same tests, such as CD4 counts to monitor the HIV infection at the same schedule as usual to monitor your infection. This will be done as part of your standard of care.

**Payment for participation**
You will not be paid for your participation in this research.

**Confidentiality of records**
Your research records will be confidential to the extent permitted by law. You will be identified by a code, and personal information from your records will not be released without your written permission. You will not be personally identified in any publication about this study.

**Questions**
If you ever have questions about this study or in case of research-related clarifications, you should contact Dr. Jessica Mulindwa at Cell phone 260 955 249 100, e-mail Jessica.mulindwa@gmail.com
The Chairperson, Biomedical Research Ethics Committee at Telephone 256 067, e-mail unzarec@unza.zm

**Legal rights**
You are not waiving any of your legal rights by signing this consent form.

**Signatures**
You will receive or have received a copy of this informed consent. Your signature below indicates that you agree to participate in this study.

| _____ | _____ | _____ |
|---|---|---|
| Signature of Participant | Printed Name of Participant | Date |
| _____ | _____ | _____ |
| Signature of Witness | Printed Name of Witness | Date |
| _____ | _____ | _____ |
| Signature of Investigator | Printed Name of Investigator | Date |
| _____ | _____ | _____ |
| Signature of Person obtaining consent (If other than investigator) | Printed Name of Person obtaining consent (If other than investigator) | Date |