ANTIRETROVIRAL DRUGS TOXICITY AND IMMUNE STATUS OF HIV PATIENTS UNDER COMPREHENSIVE CARE IN EMBU, KENYA

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ABSTRACT
The objective of this study was to determine toxicity and immune status of HIV patients under ARVs (Zidovudine, Stavudine and Nevirapine) which comprises first line ARV combination regime currently being used in Kenya. A total of sixty (60) HIV patients participated in the study after consenting to undergo comprehensive care. A control group of forty (40) HIV patients on only contrimoxale (septrin), Analysis of Variance (ANOVA), t-test and correlation coeficient were used to analyse the data. The results showed treatment with ARVs may cause . Based on the findings of this study it is recommended that the ARV drug combination used may be initiated for patients at baseline CD4 counts of 101-150 cells/µl of blood. However change to another ARV drug combination regime may be introduced after three months of use to minimize toxicity associated with prolonged use.

KEYWORDS: Antiretrovirals, Toxicity, Immune status, Creatinine

1 INTRODUCTION

Human immune deficiency virus (HIV) and acquired immune deficiency syndrome (AIDS) is a global pandemic which has reduced quality of life for millions of people (WHO, 2006). HIV is associated with immune suppression which can be measured by the levels of T cell (Roit *et al.,* 2006). Specifically HIV has been shown to reduce levels of CD4 and CD3 in addition to other haematological values (Cheeseborough *et al.,* 2005). Normal levels of CD4 in health ranges are 1000-1200 cells/µl of blood (Cheeseborough *et al.,* 2005). Individuals with or less than 200 CD4 cells/µl of blood have been shown to develop AIDS which is associated with opportunistic infections and malignancies (Gilks,1998). Currently, 40 million people are living with HIV and AIDS all over the world (WHO, 2009). It is estimated that over 1.2 million people in Kenya are infected with HIV (WHO, 2009, KAIS, 2009). Prevalence of infection in Kenya varies from region to region, with Nyanza having 14% followed by Nairobi with 9% (GOK, 2008). Embu in Eastern Province has reported a prevalence of 4% (GOK, 2008).World Health Organization has established that ARVs reduce the suffering of

HIV patients (WHO, 2006). Antiretroviral drugs have been shown to boost the immune status of HIV patients and reduce opportunistic infections (Janeway *et al*., 2005). The Kenya government has made a lot of effort to ensure that ARVs are available in provincial and district hospitals (GOK, 2006). Currently, combination ARV regimes are in use. Nucleoside analogues are most commonly used and HIV patients are initiated with first line combination ARV therapy (GOK, 2008) Antiretroviral combinations are designed to be taken for long periods of time; this could lead to emergence of ARVs associated toxicity (Smith, 2006). Toxicity can be measured by determination of levels of creatinine in serum among others which is more reliable as it indicates kidney filtration rates (Greer, 1999). Normal creatinine values in health is 0.6 to 1.2 mg/dl of blood in male adults and 0.5 to 1.1 mg/dl of blood in females and above 0.2mg/dl of blood in infants. Toxicity of most drugs may be manifested as abnormally elevated creatinine levels in serum (Greer, 1999). The objective of this study was to determine the effects of first line combination ARV therapy (nevirapine, zidovudine and stavudine) on creatinine levels and the relationship between creatinine levels and immune status of HIV patients .

2.0 METHODOLOGY

The study was conducted at Runyenjes Sub- District Hospital, Embu. The Hospital is along Embu-Meru Road 40 km from Embu Town. Runyenjes Town is at altitude 1480M above sea level, longitude 0 37o 30'E and latitude 0 3o 32'S and is the headquarters of Embu district mostly populated by Aembu with traces of other tribes. It's a transit town with many travelers from Nairobi to Meru. Embu district has an approximate population of 150,000 people and 6 thousand (4%) of these are infected with HIV (GOK, 2008). This was a longitudinal study covering HIV patients attending Runyenjes CCC during the month of may to December 2009. A representative sample of those who enrolled was randomly picked from theCCC attendance register forming the experimental group. The control group was a representative sample of HIV patients registered for opportunistic infections management. The baseline values of CD3, CD4, MCV platelets, Hb and creatinine were established. Baseline CD4 counts were used to group the patients into categories. Monitoring of creatinine, CD3, CD4, platelets, MCV and Hb was carried out monthly upto 6 months. Patients were put on first line combination ARV therapy (Nevirapine, Zidovudine and Stavudine) and monitored monthly. A control group of 40 HIV patients on contrimoxale (septrin), an antibiotic commonly used to control opportunistic infections was also monitored for creatinine, CD3, CD4, platelets, MCV and Hb monthly for six months. This was to exclude the compounding effects of accompanying antibiotics. Patients on ARVs (experimental group) were HIV patients with less than 200 CD4 cells/µl of blood at baseline while those on septrin (control group) were HIV patients with CD4 counts higher than 200 CD4 cells/µl of blood at baseline as recommended (WHO, 2006).This sample size was estimated using the formula used by Fishers *et al.* (1998) on an estimated population of 100 patients currently on ARVs in the Hospital. A sample size of 69 patients was used. The extra 10 patients were to cater for losses due to natural attrition and drug defaulters. Usually HIV patients on ARVs are given antibiotics and HIV patients with CD4 counts greater than 200 cells /µl of blood are not given ARVs but antibiotics alone. Therefore a control sample of 40 HIV patients on septrin, to control opportunistic infections was used to exclude the compounding effects of accompanying antibiotics toxicity. HIV patients attending Runyenjes CCC who consented for blood sample analysis were considered for the study. Four milliliters of venous blood was drawn from patients at every visit by a qualified clinician. The sample was divided into two aliquots of 2mls each. One of the aliquots was put into a tube containing Ethylene diamine tetra acetic acid (EDTA) anticoagulant and the other into a tube without anticoagulant. This was used for serum extraction. The sample with EDTA was used for CD3 and CD4 counts in a Fluorescent Activated cell sorter (FACs, Anaspec model) scanner using ant CD3 and ant CD4 cross-reactive monoclonal antibodies coated with

magnetic beads from USA. A sample of 20 µl of blood was mixed with coated monoclonal antibodies in a FACs machine. The results were read on a computer connected to the FACs machine as described by Cheesborough (2005). Mean corpuscular volume was measured by a blood cell analyzer by Beckam Coulter and Symex which counts cells by impendence (Cheesborough, 2005). A volume of 10µl of blood sample mixed with 100µl of buffered electrolyte was passed through an aperture tube between two electrodes. The counts were expressed in femtolitres as described earlier (WHO, 2006). Haemoglobin was measured using Haemoglobincyanide (HiCN) technique where whole blood was diluted in 1: 20 in a modified Drawkins solution containing potassium ferricyanide and potassium cyanide (Scott and Lewis, 1995). Absorbance of HiCN was read in a spectrophotometer at wavelength 540 nanometers. The absorbance obtained was compared with reference standard solution. Haemoglobin values were directly read out on the haemoglobinometer from the digital display (Scott and Lewis, 1995). Platelet count was determined by use of improved Neubauer ruled counting chamber. Blood sample was diluted in 1: 20 in a filtered solution of ammonium oxalate reagent which lyses the red cells. The platelets were then computed and expressed as number of cells per liter of blood (Pettit, 2000). Serum was used for creatinine determination in a serum analyzer. This assay is based on the reaction of creatinine with sodium picrate as described by Jeff (2008) (Labman Diagnostic UK Ltd). Picric acid (17.5 mol/l) was mixed with sodium hydroxide (0.29mol/l) and 2mls of serum loaded in the serum analyzer. The amount of creatinine was then read from the computer of the serum analyser and expressed in milligrammes per decilitre of blood (Jeff, 2008).

2.1 Data analysis

Data collected was averaged per sampling and categorized. Creatinine levels and immunological parameters were compared against the base line levels for each patient. These were grouped into four categories based on baseline CD4 levels of 1-50, 51-100, 101-150, and 151-200 (per ul of blood). Baseline CD4 counts were used to group the patients into categories so as to establish the effects of ARVs and associated toxicity when initiated at varying levels of CD4 in HIV patients. The data was plotted to show the relationship of creatinine levels following ARV administration, and relationship of creatinine with CD3, CD4, Hb, platelets and MCV following ARV therapy. The relationship between ARVs association in toxicity and immune profiles was established by use of correlation coefficient, differences between baseline values of creatinine and immune profiles and values at 6 months of ARV use was analysed using analysis of variance. Data from patients on first line combination ARV therapy (experimental group) and those on septrin (control group) was compared by t- test using paired comparison and differences between months was analysed using post ANOVA (Student Newman-Keul's test). $p<0.05$ was considered significant.

3 RESULTS AND DISCUSSION

The results showed significant difference between creatinine and immune profiles values for patients under ARVs treatment when baseline and values at 6 months was compared, with values at 6 months showing a clear elevation above baseline values (Table 4.1a: $p=0.01$, $F=22.73$, $df= 5$). In contrast, the control group, showed no significant difference between mean creatinine and immune profiles at baseline compared with mean creatinine and immune profiles at 6 months of contrimoxale use (Table 4.1b: $p=0.1$, $F=0.009$, $df= 5$).The results also showed significant differences in creatinine and immune profiles between patients under ARVs treatment and those under septrin, with patients under ARVs showing elevated creatinine and immune profiles (Table 4.1c: $p= 0.01$, $t=2.43$).

3.1 Relationship between ARV administration and CD4 and CD3 counts

In this study, the CD4 levels were determined at baseline and monthly for 6 months following ARV use in various categories of patients. The mean CD4 increased from 101 cells /µl of blood at baseline to a mean of 358 cells/µl of blood in the third month of ARV use then decreased to a mean of 278 cells/µl of blood after 6 months following ARV use. The CD4 counts increased to a peak of 358 cells/µl of blood in the third month then declined. There were significant differences of CD4 counts between the months of ARV use. CD4 counts at baseline were significantly different with CD4 counts at the third month of ARV use ($p<0.05$, $q=20.9$) , and CD4 counts at third month were significantly different with CD4 counts at sixth month of ARV use (Figure 4.1: $p <0.05$, $q=6.71$). The control group results showed no significant changes in CD4 counts following 6 months of contrimoxale (septrin) use ($p=0.1$, $F=0.003$, $df=5$). After contrimoxale treatment mean CD4 counts decreased from 250 at baseline to 209 cells/µl of blood at 6 months of contrimoxale use. Among the various categories, patients with baseline CD4 counts of 100-150 cells /µl of blood showed a better and highly significant response with a mean of 405 cells /µ l of blood at the 6 months follow up than all other categories. This showed that, use of ARVs resulted in elevated levels of mean CD4 counts especially for patients who started treatment when CD4 counts were 100-150 cells/µl of blood (Figure 4.2; $p=0.01$, $F=15.2$, $df=3$). The CD3 level was determined at baseline and sequentially for 6 months following ARV use in different patient categories based on CD4 baseline counts. The results revealed that mean CD3 increased from 1306 cell/µl of blood at baseline to 1778 cells/µl of blood following 6 months of ARV use. This showed that use of ARVs for a long time resulted in elevated levels of CD3 counts to a certain level which may then stabilize (Figure 4.3). Patient category with baseline CD4 of 101-150 cells/µl of blood showed the best CD3 response (Table 4.3). The control group results showed no significant changes in CD3 counts following 6 months of contrimoxale (septrin) use ($p=0.1$, $F=0.12$, $df=5$). After treatment with contrimoxale mean CD3 counts decreased from 1600 cells/µl of blood at baseline to 1237 cells/µl of blood at 6 months. This showed that contrimoxale use may not improve CD3 counts (Figure 4.3).

3.1.1 Effects of ARVs (Zidovudine, Stavudine and Nevirapine) on CD4 and CD3 Counts

The normal range of CD4 is 1000-1200 cells/µl of blood in a healthy person (Janeway *et al.,* 2005). In this study CD4 counts increased from mean values of 101cells/µl of blood at baseline to 358 cells/µl of blood in the third month of ARV chemotherapy, followed by a decline to 278 cells/µl of blood in the sixth month of chemotherapy. Similar studies by Uberg, (2009) which reported that, CD4 counts increase rapidly in the initial levels of ARV therapy then decline, may be due toxic pressure of ARVs. Studies by Antoni *et al.* (2002) showed that the toxicity of ARVs on the bone marrow and blood-cell system could result in re-infection with HIV. The control group showed no significant changes in CD4 counts with a mean of 231 cells/µl of blood following 6 months treatment with contrimoxale for opportunistic infection management. This showed that septrin had no significant effect on CD4 levels. CD3 counts increased from baseline mean values of 1306 cells/µl of blood to a mean of 1887 cells/µl of blood in the fourth month of ARV chemotherapy followed by a decline to 1778 cells/µl of blood in the sixth month of chemotherapy. The peak mean CD3 count of 1887 cells/µl of blood was observed in the fourth month of treatment then declined to 1778 cells/µl of blood in the sixth month corresponding with decreasing CD4 counts. T cells response is crucial in all intracellular infections like HIV. CD3 is a marker of all T cells and its changes indicate levels of cellular immune responses (Roit *et al.,* 2006). CD4 T cell response induces both humoral and cellular response but also crucial as a major receptor of HIV entry. Hence changes in CD4counts could result changes in CD3 counts. This concurs with results

attained by Carbonara *et al.* (2001) who found an increase in CD3 levels with increase in CD4 counts and vise versa.

3.2 Relationship between ARV administration and creatinine levels

To assess toxicity, the mean creatinine levels were determined at baseline and 6 months following ARV use in various categories of patients. The results revealed significant differences in mean creatinine levels which increased from 1.03 mg/dl of blood at baseline to 1.37 cells/ul of blood at 6 months following ARV use. (Figure 4.4; p=0.01, F=22.73, df=5). The normal creatinine in health is 0.6-1.2 mg/dl of blood and therefore the results showed that prolonged use of ARVs leads to elevated levels of creatinine which is indicative of development of toxicity.The control group (patients on septrin) showed no significant changes in creatinine levels with a mean of 0.99 mg/dl of blood following 6 months of contrimoxale (septrin) treatment. This is within the normal range of creatinine in health. Creatinine levels changed from mean baseline values of 0.91 to 0.99mg/dl of blood at 6 months of contrimoxale treatment. However these changes were not significant (p=0.1, F=0.004, df=5). Among the patient categories there were significant differences in creatinine levels. Patients with a baseline CD4 counts of 101-150 cells/µl of blood had a better response to ARVs treatment with a mean creatinine of 1.08 mg/dl of blood compared to other categories. Patients with baseline CD4 counts of 50-100 cells /ul of blood had mean creatinine of 1.25 mg/dl of blood which was significant when compared to baseline levels (Figure 4.5; p=0.01, F=5.66, df=3). This category of patients had least CD4 response.

3.2.1 Effects of ARVs (zidovudine, stavudine and nevirapine) on creatinine levels in HIV patients

In health normal creatinine ranges from 0.6-1.2 mg/dl of blood. Creatinine level than 1.2 mg/dl of blood indicates poor kidney glomelural filtration rates which would be a sign of kidney impairement. In this study, all the patients were given fixed doses of ARVs. Creatinine levels increased with continued chemotherapy from mean of 1.03 mg/dl at baseline to 1.37mg/dl of blood at the sixth month of ARV treatment. The results showed that prolonged use of ARVs may lead to elevated levels of creatinine indicating potential of toxicity in HIV patients. The increase in creatinine was an indication that ARVs at some level impair kidney function hence reduce glomelura filtration rates resulting in elevation of creatinine in serum. A Similar observation was reported by Hirsch and Gunthad, (2005) which showed that impaired kidneys lead to increased serum creatinine. The control group which received contrimoxale for the management of opportunistic infection showed no significant changes in creatinine with a baseline mean of 0.91 mg/dl of blood and changed to of 0.99 mg/dl of blood following 6 months of treatment. This showed that contrimoxale had no significant effect on creatinine levels since this was within normal creatinine range of 0.6-1.2 mg/dl of blood.

3.3 Relationship between creatinine and CD4 counts

The mean CD4 counts were compared with mean creatinine for all the patients from baseline to 6 months of follow up during ARV administration. Creatinine was found to be positively correlated with CD4 counts upto three months of ARV treatment (Figure 4.6; $p<0.01$; $r=0.178$).

3.4 Relationship between ARV administration and haemoglobin levels

The haemoglobin (Hb) levels were determined at baseline and sequentially for 6 months following ARV use and in various categories of patients to determine the effect of ARVs on Hb. The results revealed mean Hb increased from 11mg/dl of blood at baseline to 13 mg/dl of blood following 6 months of ARV use which was within normal range. Hence Hb had no significant differences during ARV use (p=0.1, F=0.09, df =5). The

control group results showed no significant changes (p=0.5, F=0.007, df=5) in Hb levels with a mean of 12mg/dl of blood at 6 months of (septrin) treatment. The mean Hb level increased from 9.0 at baseline to 12 mg/dl of blood at six months of contrimoxale treatment (Figure 4.7).

3..4.1 Relationship between creatinine and haemoglobin levels

The mean Hb levels were compared with mean creatinine of all the patients from baseline to all the levels of ARV administration. Creatinine was found to be weakly correlated with haemoglobin (Figure 4.8; $p<0.01$; $r=0.0061$).

3.5 Relationship between ARV administration and MCV Levels

The MCV levels were determined at baseline and 6 months following ARV use in various categories of patients. The mean MCV level increased signinficantly from 88 to 104fl at six months of treatment (p=0.01, F=5.10, df=5). This showed that use of ARVs for a long time may result in elevated levels of MCV levels. The control group showed no significant changes (p=0.01, F=0.002, df=5) in MCV levels following contrimoxale (septrin) use for 6 months. The mean MCV levels changed from baseline mean of 88fl to 84 fl at 6 months (Figure 4.9).

3.5.1Relationship between creatinine and MCV Levels

The mean MCV levels were compared with mean creatinine for all the patients from baseline to 6 months of ARV administration. Creatinine was found to be positively correlated with MCV levels (Figure 4.10; $p<0.01$; $r=0.149$).

3.6 Relationship between ARV administration and platelet counts

The platelet levels were determined form baseline to 6 months following ARV use in the various HIV categories. The results revealed significant differences (p=0.01, F=9.34, df=5) with mean platelets increasing from 300 x 10^9 cells/l of blood at baseline to a mean of 506 x10^9 cells/l of blood 6 months following ARV use (Figure 4.12). This showed that use of ARVs may result to elevated levels of platelets counts. The control group results showed no significant rise (Figure 4.11 : p=0.07, F=0.006, df=5) in platelets counts levels with a mean of 150 x10^9 cells /l of blood at baseline that changed to 235 x10^9 cells /l of blood following 6 months of contrimoxale treatment.

3.6.1Relationship between creatinine and platelet levels

The mean platelet counts were compared with mean creatinine of all the patients from baseline to 6 months of ARV administration. Creatinine was found to be positively correlated with platelets (Figure 4.12; $p<0.01$; $r=0.082$).

3.7 Effects of ARVs (Zidovudine, Stavudine And Nevirapine) on Haematological values

The normal range of haemoglobin in health is usually 11-18mg/l of blood (Janeway *et al.,* 2005). In this current study, haemoglobin levels increased from baseline mean values of 11.0mg/l of blood to 13 mg/l of blood in the sixth month of ARV chemotherapy. The mean Hb observed was within the normal range in health. The results showed that treatment with ARVs resulted in increased Hb levels but within normal range. May be the level of toxicity generated was not high enough to influence Hb negatively to change out of normal range. The control group results showed no significant changes in Hb levels with a mean of 12 mg/dl of blood following 6 months

of contrimoxale use. This showed that contrimoxale had no significant effect on Hb levels. Platelet counts increased from baseline mean values of 300 x 10^9 cells/l of blood and increased to a mean of 506 x 10^9 cells/l of blood in the sixth month of ARV chemotherapy. The mean platelets counts observed after 6 months of ARV use were slightly beyond the normal range in health which is usually 150-450 x 10^9 cells /l of blood. This disagrees with results of studies done by Smith (2006) who reported that most drugs are associated with platelet destruction. Probably platelet elevation was responding to ARV drugs in circulation and the level of drug toxicity observed in this study was not high enough to cause platelet destruction. The control group showed no significant changes in platelets counts levels with a mean of 235 x10^9 cells /l of blood following 6 months of contrimoxale use. This showed that septrin may not have negative effects on platelet levels. Mean corpuscular volumes is a measure of the average red blood cell volume that is reported as part of a standard complete blood counts (Taften, 2007). Patients may indicate microcytic anaemia (MCV below normal range), normocytic anaemia (MCV within normal range) or macrocytic anaemia (MCV above normal range) (Fleischer, 2004). MCV levels increased from baseline mean values of 88 fl to a mean of 104 fl in the sixth month of ARV chemotherapy. The mean MCV observed were beyond the normal range in health which is usually 80-100 (fl). May be toxicity experienced induced elevation of MCV. This is in agreement with results of studies by Grohskopf and Black *(*2005) which reported that, ARVs such as Nevirapine, is associated with macrocytic anaemia due to increase in red cell volume in response to the drug . The control group showed no significant changes in MCV levels with a mean of 84 fl following 6 months of contrimoxale use. This showed that contrimoxale had no significant effect on MCV levels as it was less toxic.

3.8 Relationship between creatinine and immune status of HIV patients on ARVs (zidovudine, stavudine and nevirapine)

The mean CD4 counts and creatinine levels of patients under ARVs were compared from baseline to 6 months of follow up during. Creatinine was found to be positively correlated with CD4 counts for up to the third month of follow up. The mean creatinine observed in the third month of treatment was 1.18mg/dl of blood and indicated the peak mean CD4 count of 358 cells/µl of blood. Creatinine above 1.18mg/dl/of/blood, was corresponded with mean CD4 counts decreasing rapidly from mean of 358 cells/ul of blood in the third month to a mean of 278 cells/µl of blood in the sixth month of treatment. This could have been due to toxic effects of ARVs increasing beyond tolerance and affecting the blood cell system. This is supported by results of Antoni *et al.* (2002)) which showed that the toxicity of ARVs on the bone marrow and blood-cell system decreases CD4 levels. In this study, decrease in CD4 may be explained by increased toxicity as a result of ARVs prolonged administration. This showed that only creatinine beyond a certain level can indicate ARV toxicity. Patients with baseline CD4 counts of 50-100 cells /µl of blood had highest creatinine profiles of 1.25 mg/dl of blood. This was associated with lowest CD4 response of 205 cells /µl of blood to ARVs treatment while patients with baseline CD4 counts of 101-150 cells /µl of blood had the lowest creatinine profiles of 1.08 mg/dl of blood with the highest CD4 response of 405 cells /µl of blood to ARVs treatment following 6 months of ARV use. This suggest that when toxic pressure is experienced, the CD4 response is poor. The control group showed no significant changes in CD4 counts and creatinine levels with CD4 mean of 209 cells/µl of blood and mean creatinine of 0.99 mg/dl of blood following 6 months of treatment with contrimoxale. The creatinine observed in the third month of treatment was 1.01mg/dl of blood while mean CD4 count was 249 cells/µl of blood. Creatinine was within the normal range with no significant changes while CD4 counts were still low. This may suggest that viral load was still high and hence CD4 response was poor even after administration of contrimoxale (WHO, 2006). The mean Hb levels were compared with mean creatinine of all the patients from

baseline up to 6 months of ARV administration. Creatinine was found to be weakly positively correlated with haemoglobin. The highest mean creatinine observed after 6 months of treatment was 1.37mg/dl of blood and a peak Hb level of 13mg/dl of blood. Studies by Kuhn (2001) showed that haemoglobin levels may not be dependent on drug toxicity but may depend on nutritional factors. Among the patient categories those with baseline CD4 counts of 101-150 cells /µl of blood had the lowest creatinine profiles after ARVs treatment. They had a creatinine mean of 1.08 mg/dl of blood corresponding with highest Hb with a mean of 12.4 mg/dl of blood. Patients with baseline CD4 counts of 50-100 cells /µl of blood had highest creatinine profiles after 6 months of ARVs treatment with a mean of 1.25 mg/dl of blood and the lowest Hb with a mean of 11.5 mg/dl of blood following 6 months of ARV treatment. These results showed that in all categories Hb was within the normal range probably because toxicity generated was not high enough to have a negative effect on Hb. The control group results showed no significant changes in Hb levels and creatinine levels with. Hb changed from 11 mg/dl of blood at baseline to 13 mg/dl of blood after 6 months while creatinine changed from 0.91 at baseline to 0.99 mg/dl of blood following 6 months treatment with contrimoxale. The mean MCV levels were compared with mean creatinine for all the patients from baseline up to 6 months of ARV administration. Creatinine was found to be highly positively correlated with MCV levels. The highest mean creatinine observed in 6 months of treatment was 1.37mg/dl of blood and indicated the peak mean MCV level of 104 fl. This is in agreement with results of studies by Hirsch and Gunthad (2005) which reported that, increased drug toxicity may increase MCV above normal range resulting from increased red cell volumes.

Among the patient categories those with baseline CD4 counts of 101-150 cells /µl of blood had their creatinine and MCVprofiles least affected by ARVs treatment. With a creatinine of 1.08 mg/dl of blood and MCV of 98.5 fl. Patients with CD4 counts of 50-100 cells /µl of blood had their creatinine profiles greatly influenced by ARVs treatment with creatinine of 1.25 mg/dl of blood and MCV of 107.7 fl after 6 months of ARV treatment. The control group showed no significant changes in MCV levels and creatinine levels. MCV changed from 88fl at baseline to 84fl while creatinine changed from 0.91fl to 0.99 mg/dl of blood after 6 months of contrimoxale use.The mean platelet counts were compared with mean creatinine of all the patients from baseline up to 6 months of ARV administration. Creatinine was found to be positively correlated with platelets. The highest mean creatinine observed in 6 months of treatment was 1.37mg/dl of blood and indicated the peak mean platelets level of 506 x 10^9 cells /l of blood. This is in agreement with results of studies done by Carbonara and David (2001) which reported that, ARVs toxicity is associated with thrombocytosis. Thrombocytosis is associated with elevated platelets levels in serum.Among the patient categories, patients with baseline CD4 counts of 50-100 cells /µl of blood highest creatinine profiles with a mean of 1.25 mg/dl of blood corresponding with highest platelets of 488 x 10^9 cells /l of blood. Patients with CD4 counts of 101-150 cells /µl of blood had the least creatinine profiles of 1.08mg/dl of blood and lowest platelets of 389 x 10^9 cells /l of blood. The control group showed no significant changes in platelets levels and Creatinine levels changed from 0.91 at baseline to 0.99 mg/dl of blood following 6 months of contrimoxale use.

In this study, it was observed that the mean creatinine and mean immune profiles (MCV, platelets and Hb attained in the third month were within the normal ranges in health except CD4 counts which were far below normal. This could be due to the fact that at this level of ARV therapy, it is possible that there was gradual restoration of the immune system as indicated by the rising CD4 counts. It may also be suggested that normal CD4 levels in health may not be easily attained in HIV patients under ARV therapy. Probably drugs suppress body cells proliferation and destroy body cells (Smith, 2006). Beyond three months of ARV treatment of HIV patients, ARV drug may have impaired the immune system leading to impaired immune responses.

The results of the control group showed no significant changes in creatinine levels and immune responses with mean creatinine of 0.994 mg/dl of blood following 6 months treatment with septrin. The mean creatinine observed in the third month of treatment was 1.01mg/dl of blood and indicated mean CD4 count of 249 cells/ul of blood. Creatinine was within the normal range with insignificant changes while CD4 counts were still low. This showed that contrimoxale toxicity had no significant effect on the immune responses.The current study showed that serum creatinine levels beyond 1.18mg/dl of blood may be a good indicator of ARV drug toxicity. This level may be used by clinicians to change the ARV drug regime to boost the immune system in management of HIV patients. The study also show that ARV chemotherapy could be started at baseline CD4 counts of 101-150 cell/ul of blood because this is when there is least ARV associated negative impact on the immune responses based on observed creatinine levels. Creatinine levels in serum may be used as an indicator of ARV toxicity in HIV patients.

ACKNOWLEDGEMENTS

I wish to sincerely thank all the staff of Runyenjes hospital for their support especially Dr. James Maina and Dr. Elesban Kihuba who allowed me through the ministry of medical services to carry out this study at Runyenjes comprehensive care centre, Mrs. Anceta Njeru, the incharge of the comprehensive care centre who assisted me to bleed patients for blood samples, the Embu provincial laboratory staff especially Mr. John Munyi who assited me to analyse the blood samples with the laboratory equipments. My gratitude also goes to Kenyatta University for giving me an opportunity to to be their student. My sincere thanks also go to Peter Njeru of Karurumo health centre for his encouragement as I carried out this study. Last but not the least I am grateful to my family especially my father Samuel Kiura for his financial support and friends for their unlimited moral support.

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Table 4.1a: Mean creatinine and immune profiles of HIV patients at baseline and 6 months of ARV treatment

Profiles	Creatinine	CD4	CD3	Hb	Platelets	MCV
Baseline	1.03	101	1306	11	300	88
6 months	1.37	278	1778	13	506	104

Creatinine (mg/dl of blood), CD4 and CD3 (cells/µl of blood), Hb= Haemoglobin (mg/dl of blood), platelets (cellsx10^{9}/l of blood), MCV=Mean corpurscular volume (femtolitres)

Table 4.1a: Mean creatinine and immune profiles of HIV patients at baseline and 6 months of ARV treatment

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Table 4.1b: Mean creatinine and immune status of HIV patients at baseline and 6 months of contrimoxale use

Profiles	creatinine	CD4	CD3	Hb	Platelets	MCV
Baseline	0.91	250	1600	9.0	150	88
6 months	0.99	209	1237	12	235	84

Table 4.1c: Mean creatinine and immune profiles of HIV patients on ARVs and HIV patients on contrimoxale (septrin) at 6 months of treatment

Profiles	Creatinine	CD4	CD3	Hb	Platelets	MCV
Patients on ARVs	1.37	278	1778	13	506	104
Patients on septrin	0.99	209	1237	12	235	84

Table 4.2: Patients categories based on baseline CD4 levels and their corresponding mean creatinine and immune profiles 6 months of ARV use

CD4 count (category)	No. of patients(%)	creatinine	CD4	CD3	Hb	platelets	MCV
<50	20 (33.3%)	1.18	205	1544	12.1	404.9	100.2
50 -100	9 (15%)	1.25	196	1461	11.5	488	107.7
101-150	8 (13.3%)	1.08	405	2054	12.4	389	98.5
>150	23 (38.4%)	1.21	319	1738	12.3	457	100.9

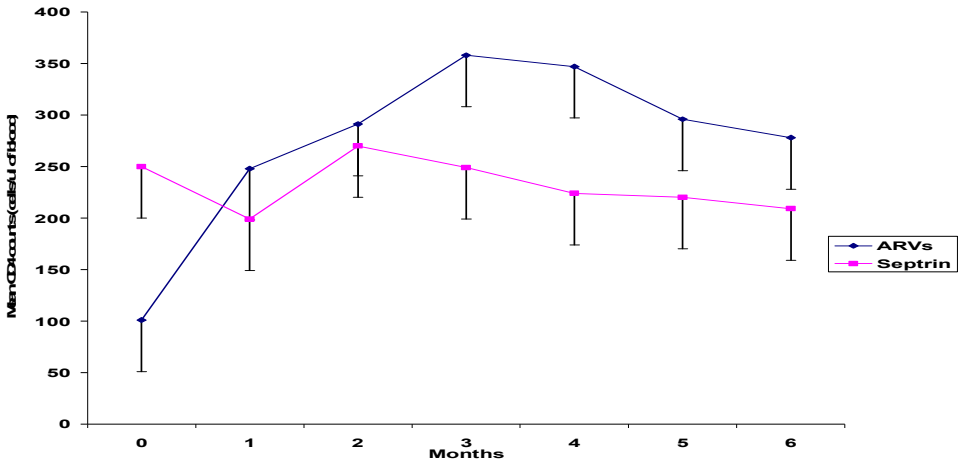

Figure 4.1: Mean CD4 counts and sequential counts 6 months following ARV and septrin use respectively

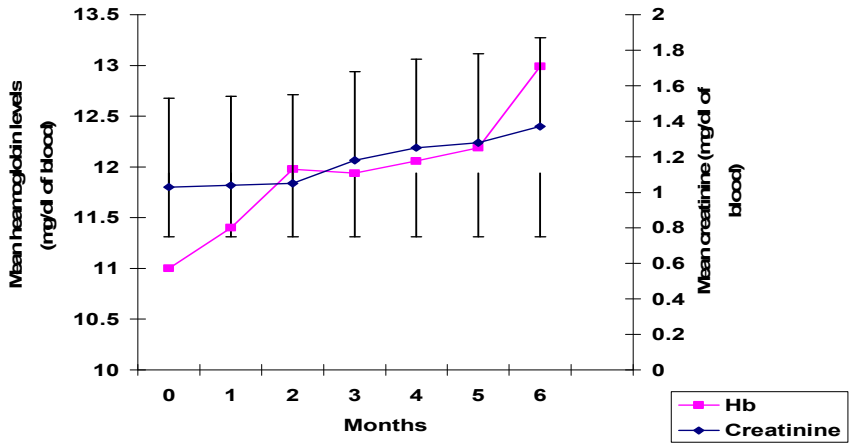

Figure 4.8: Relationship between mean heamoglobin levels and mean creatinine values 6 months of ARV use

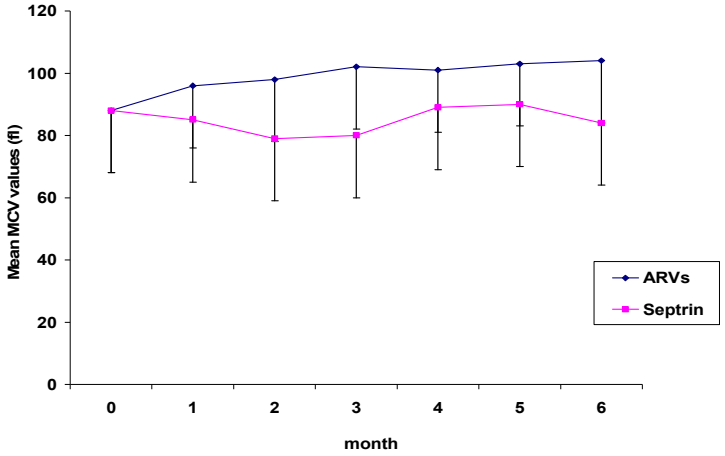

Figure 4.9: Mean MCV baseline values and sequential values 6 months of ARV and septrin use respectively

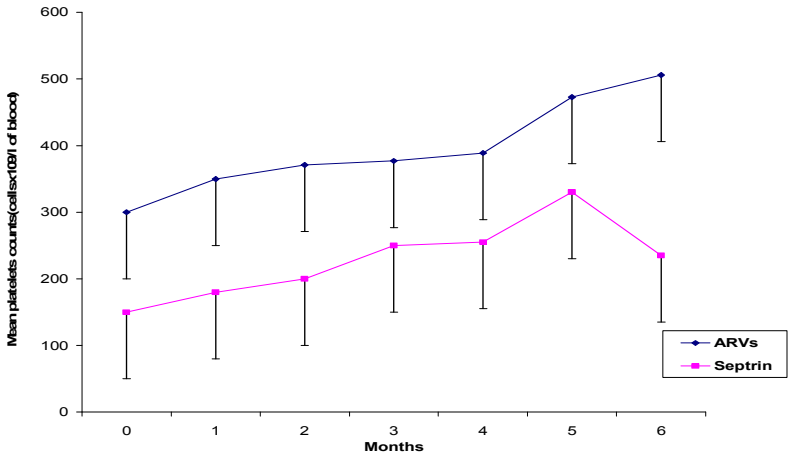

Figure 4.11: Mean platelets counts and sequential counts 6 months of ARV and septrin use

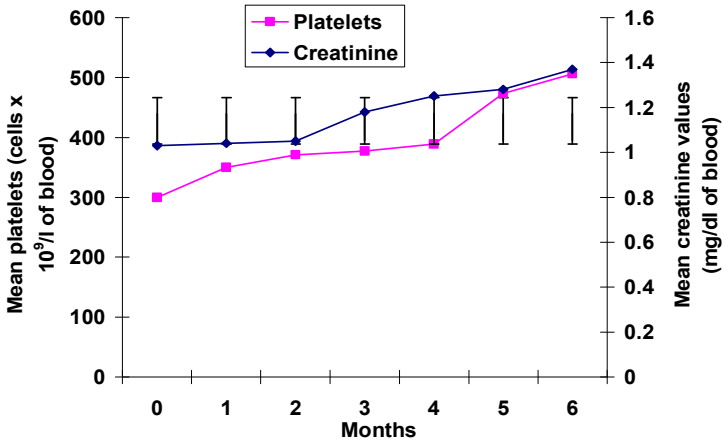

Figure 4.12: Relationship between mean platelets and mean creatinine levels at baseline and subsequent levels for 6 months of ARV use