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INCIDENCE OF ANAEMIA AMONG HIV-INFECTED PATIENTS TREATED WITH ZIDOVUDINE-CONTAINING ANTIRETROVIRAL THERAPY IN NORTHEASTERN NIGERIA

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ABSTRACT

Background: **Zidovudine (AZT) is a common component of antiretroviral therapy (ART) in resource-limited settings. However, AZT is associated with myelotoxity that often presents with anaemia. The aim of this study was to determine the incidence of anaemia among patients initiated on AZT-containing and non-AZT containing ART regimens.**

Methods: **In this retrospective analysis, records from 800 ART-naïve HIV-infected patients were abstracted by simple random sampling from program databases. Rates of anaemia were compared between patients initiated on AZT- versus non-AZT-containing ART regimens. Patients were stratified according to absence (Group A) or presence (Group B) of baseline anaemia defined as haemoglobin < 10.5g/dl. Incidence was calculated as total cases of AZT-induced anaemia (group A) or worsening of anaemia (group B) during the study period divided by person-time at risk and adjusted per 100 person-years. Average time-to-event and survival curve were estimated using Kaplan Meier survival analysis.**

Results: **In group A (without baseline anaemia), the incidence of anaemia in the AZT-exposed versus non-exposed cohorts was 73.3 and 17.6 per 100 patient years at 6 months, and 60.5 and 8.5 per 100 patient years at 12 months, respectively. In group B, the incidence of worsening anaemia was 65.9 and 26.2 per 100 patient years at 6 months, and 57.5 and 17.9 per 100 patient years after 12 months in AZT-exposed and AZT-unexposed cohorts, respectively. The estimated time to event (developing anaemia) was 2.3 (1.5 – 3.4) months, while estimated to event (worseninig anaemia) was 2.0 (1.5 – 4.0).**

Conclusions: **HIV-infected patients initiated on AZT-containing ART are 2.7 and 4.5 more likely to develop anaemia at 6 and 12 months, respectively, compared to those initiating a non-AZT containing regimen. When censored at 12 months the overall incidence of AZT-related anaemia was estimated at 22.3% (38.2 incidences per person years). Majority (75%) of the AZT-related anaemia occurred early with estimated time-to-event occurring within the first 3.8 months**

Keywords: Anaemia, Zidovudine, Antiretroviral therapy, Incidence

INTRODUCTION

Anemia is a common hematological manifestation of HIV infection associated with mortality especially among those with advanced disease.[1] Studies that estimate the prevalence of anemia among HIV infected persons in resource limited nations that bear the greatest burden of the disease indicate that it ranges from 40% to 90%.[2] Several factors including stage of HIV, age, sex, ART regimen, cut off value for the definition of anemia, and geographical location contribute to the variation in rates of anaemia.[2] Zidovudine (AZT), a thymidine nucleoside analog reverse transcriptase inhibitor (NRTI), is effective in the management of HIV infection. The projected market share of AZT for adults as a proportion of NRTI stood at 20% as of 2016 in most low and middle income countries.[3] The WHO recommends use of tenofovir (TDF) as a component of first-line ART instead of AZT given the associated increased risk of anemia.[4] AZT-induced anaemia often occurs soon after initiating treatment and this haematologic toxicity is thought to be dose-dependent. Further, AZT-induced anemia has been shown to increase the risk of mortality and morbidity and adversely affect quality of life of HIV-infected persons.[5] Studies have established an association between anemia at baseline and decreased survival/increased disease progression in patients with

HIV.[3-5]. AZT-induced anaemia could result from bone marrow depression, described as proliferative inhibition of red cell progenitors[6] or reversible pure red cell aplasia (PRCA).[7] In the Development of Antiretroviral Therapy in Africa (DART) study in Uganda, it was observed that 20% of patients developed new anaemia after initiation of AZT-containing ART, with 5% developing severe anaemia (hemoglobin level <6.5 g/dL); 20% of the anaemia was classified as macrocytic and thus presumably AZT-related.[8] Yet most of the studies that evaluated AZT-induced anaemia were conducted in Western countries and may not reflect findings from developing nations. There is a dearth of research from sub-Saharan Africa that has the highest burden of HIV infection. To the best of our knowledge, there are scarce published studies on AZT-induced anaemia in Nigeria, particularly the north-eastern part of the country. We therefore evaluated the incidence of anaemia among HIV-infected patients treated with AZT-containing ART in northeastern Nigeria.

METHODS

This retrospective study was conducted at the University of Maiduguri Teaching Hospital (UMTH). Participants included HIV-infected adults and adolescents who were initiated on ART from 2nd January 2010 to 30th December 2011. The UMTH is a 530-bed hospital, designated Centre of Excellence for Infectious Diseases, that provides tertiary health care services to individuals in north-eastern Nigeria and the neighboring countries of Cameroun, Chad, and Niger Republics.

Inclusion criteria

- Non-pregnant HIV-infected adults and adolescents (i.e. >15 years old).
- Commenced ART for the first time in his/her life within the period of study at UMTH (i.e. ART-naïve at commencement).
- Available baseline laboratory values (specifically haemoglobin level)

Exclusion criteria

- Pregnant women
- Patients ≤15 years of age
- Subjects without any follow-up data

Eligible population for this study, considering the above inclusion and exclusion criteria, was 1910 patients.

Censoring

Censoring of subjects during the study period was determined by the following criteria:

- Development of anaemia during the study
- Switch of antiretroviral therapy
- Death
- Lost-to-follow-up
- End of the research

Sample size

Sample size was estimated using the formula:

$$n = \frac{(z_\alpha + z_\beta)^2 p(1-p)}{d^2}$$ (Ibrahim, 2009)[9]

Where:

n= desired sample size for each of the cohort group in an infinite sampling frame

z_α= normal standard deviate (at 0.05 a-level, the corresponding value of z is 1.96)

z_β= normal standard deviate (at 80% power, the corresponding value of z is 0.84)

p= Incidence of AZT-induced anaemia, considered 15% (0.15) based on literature review

d= expected difference (0.05 would be considered at 95% CI)

$$n = \frac{(1.96+0.84)^2 * 0.15(1-0.15)}{(0.05)^2} = 399.8$$

Thus, a sample size of 400 patients was required for each group, including those receiving AZT-containing and non-AZT containing regimens, in order to achieve 80% power with an alpha of 0.05. A total of 800 patient records were therefore required to study the population.

Sampling techniques

Random sampling of electronic medical records was employed to select the necessary sample size for each group from their respective sampling frame. After using both the inclusion and exclusion criteria to filter the adult and adolescent databases, the final eligible subjects were divided into two groups based on their initial ART regimen (AZT-containing ART (1003) and non-AZT containing ART (907)). Each group was serially arranged and assigned sequential numbers. A total of eight hundred (800) subjects (i.e. 400 subjects from each of the group) were randomly selected using the random number generator component of Statistical Package for Social Sciences (SPSS) version 20.0). This process of sampling is explained by the flow diagram below:

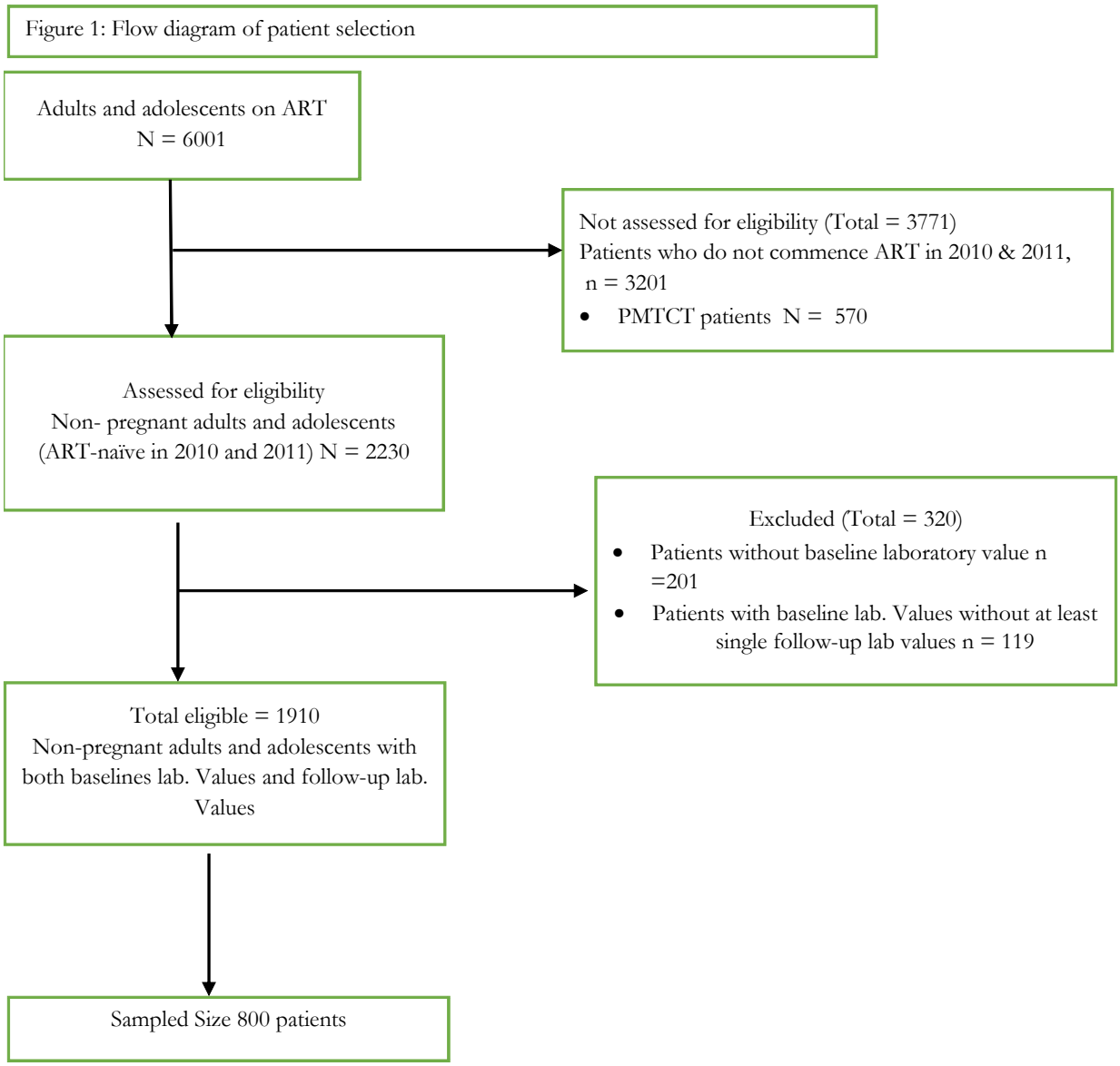

Fig.1: Flow diagram of the cohort enrolled

Data collection:
Information abstracted from the database includes age, gender, weight, haemoglobin concentration, CD4 cell count and HIV viral load. Other information includes use of co-trimoxazole as prophylaxis and ART regimen.

Operational Definitions
Anaemia-of-all-causes: defined as a haemoglobin concentration less than or equal to 10.5 gm/dl for both male and female.
AZT-related anaemia: defined as all cases of reported AZT toxicity that were documented in either toxicity database or on the comment section of the pharmacy database for subjects enrolled for the study.

Worsening anaemia: defined in this context as reduction in haemoglobin concentration by greater than or equal 1 gm/dl for subjects with baseline anaemia
Severe anaemia: defined using WHO anaemia grading as a haemoglobin concentration less than 6.5 g/dl.
Thrombocytopenia: defined as platelet count of less than 150,000/ìL.
Neutropenia: defined as absolute neutrophil count (ANC) of less than1000cell/ml.
Renal impairment: Defined as estimated creatinine clearance (CrCl) less than 60 ml/min. The CrCl was estimated using Cockcroft-Gault formula: for male $estimate_{CrCl} = \frac{\text{using Cockcroft-G}}{0.81} \frac{(140 - Age)wt}{*\ Serum\ creatinine\ (SCSCr)}$ and multiplied by 85% for female. The serum creatinine was in µmol/l.
Lost-to-follow: Defined as subjects with less than six months follow-up data available.
Time-to-event (anaemia or worsening of anaemia): Defined as the midpoint of the time interval between being case free and becoming a case.

Data analysis

The abstracted data for the 800 patients initiated on AZT-containing (n=400) and non-AZT containing (n=400) ART regimens were exported to SPSS version 20.0 via Excel spread sheet and analyzed.

Qualitative data analysis techniques employed include simple description like frequency and percentages. Incidence was calculated as total cases of AZT-induced anaemia or worsening of anaemia during the study period divided by person-time at risk and adjusted per 100 person-years. Normality tests were performed for continuous variables using a one-sample Kolmogorov Smirnov test. Statistical mean of normally distributed variable was compared using two-sample independent Student's t-test and median of non-normally distributed variables were compared using Mann-Whitney U test across dichotomous categorical variables. Average time-to-event and survival curve were estimated using Kaplan-Meier survival analysis. Inferential analyses were performed at 95% confidence interval (CI) and a p-value <0.05 was considered to be statistically significant.

RESULTS

Study population

The 800 study participants were stratified into two groups based on baseline anaemia status: Group A consisted of patients with no baseline anaemia, and Group B of those with baseline anaemia. Each group was further divided into two sub-groups: those initiated on non-AZT containing ART without and with baseline anemia were sub-grouped as A1 and B1, and those initiated on AZT-containing ART were sub-grouped as A2 and B2, respectively. Within group A, those that were initiated on non-AZT containing ART regimens (subgroup A1) were younger, consisted of more females, had higher CD4 cell counts, lower mean haemoglobin concentrations, greater proportion receiving cotrimoxazole prophylaxis, lower proportion of participants with AIDS (immunological or clinical) or advanced HIV infection (immunological or clinical), fewer with weight less than 50 kg, and fewer with renal impairment than those in subgroup A2, who received AZT-containing ART regimen. Median weight, median HIV viral load, hepatitis C infection, hepatitis B virus (HBV) infection, incidence of thrombocytopenia and neutropenia were comparable between subgroups A1 and A2.

Among patients in group B, with baseline anaemia participants in subgroup B2 were younger, had higher mean haemoglobin concentrations, more received cotrimoxazole prophylaxis, fewer had neutropenia and were less likely to have AIDS (clinical), advanced HIV infection (clinical), HBV coinfection, thrombocytopenia, or renal impairment than subgroup B1. Median weight, median viral load, proportion with weight less than 50 kg, HCV coinfection, CD4 cell counts, AIDS (immunological), advanced HIV infection (immunological), female proportion and those that were lost to follow-up were comparable between subgroup B1 (initiated on AZT -naïve regimen), and B2 (initiated on AZT). The baseline characteristics of participants are presented in Table 1.

Table 1(a): Characteristics of participants on AZT and non- AZT stratified by baseline anaemia

	Group A Patient without baseline anaemia HB±SD = 12.101 ±1.1096			Group B Patient with baseline anaemia HB±SD = 8.738 ±1.264		
	Group A1 Non-AZT (n=177)	Group A2 AZT (n=225)	p-value	Group B1 Non-AZT (n=223)	Group B2 AZT (n= 175)	p-value
↕Gender: Female	70 (38.4)	148 (65.8)	< 0.001	139 (62.3)	120 (68.6)	0.195
↕Weight (< 50kg)	28 (16.4)	29 (13.8)	0.485	84 (40.2)	56 (32.6)	0.124
CD4 Cell count <200 cells/ul (Immunological AIDS)	116 (65.5)	126 (49.3)	0.001	156 (69.5)	110 (62.9)	0.163
↕Advanced HIV infection (WHO stages III&IV)	176 (97.7)	210 (93.3)	0.040	218(98.2)	169 (96.6)	0.304
↕Cotrimoxazole use	58 (32.8)	150 (66.7)	< 0.001	82 (36.8)	114 (65.1)	< 0.001*
↕HCV infection	0 (0.0)	203 (0.9)	0.210	199 (0.9)	3 (1.7)	0.471
↕HBV infection	24 (13.6)	27 (12.9)	0.826	33 (14.9)	8 (4.6)	0.001*
↕Neutropenia	26 (14.7)	47 (21.1)	0.100	18 (8.1)	25 (14.5)	0.042*
↕Thrombocytopenia	30 (1.7)	61 (2.7)	0.513	13 (5.8)	2 (1.1)	0.015*
↕Renal impairment	33 (18.7)	15 (7.1)	0.001	100 (47.8)	49 (28.5)	< 0.001*
↕Lost-to-follow	28 (15.8)	44 (19.6)	0.332	48 (21.5)	42 (24.0)	0.558

*HB±SD = Mean haemoglobin concentration ± Standard deviation. *significant at p value < 0.05.*

Table 1(b): Mean ages and immunovirological parameters stratified by baseline anaemia.

	Group A Patients without baseline anaemia			Group B Patients with baseline anaemia		
	Non-AZT	AZT	Pvalue	Non-AZT	AZT	P value
†Age (years) median (IQR)	36 (28-42)	31 (26-38)	0.001*	34 (27-42)	32 (26-40)	0.177
†Weight(kg) median (IQR)	60 (52-67)	59.25 (51.9-65.0)	0.403	52.5 (46.0-61.0)	53 (47.0-62.0)	0.176
†CD4+ T lymphocyte countmedian (IQR)	158 (102-248.5)	201 (117.0-265.5)	0.031*	142 (66-242)	155 (59-244)	0.466
†Viral load median (IQR)	49,107 (5430-242,467)	31,346 (2,044.0 - 143,445.0)	0.079	49,838 (5,687-291,834)	51,333 (5,929–179,762)	0.684
‡Hb Conc. (mean (SD))	12.32 (1.3)	11.93 (0.8)	0.001*	8.41 (1.3)	9.16 (1.0)	<0.001

**significant at p value < 0.05*

↕X^2-test employed to compare proportions

† Mann-Whitney U employed to compare median

‡Independent student t-test employed to compare mean

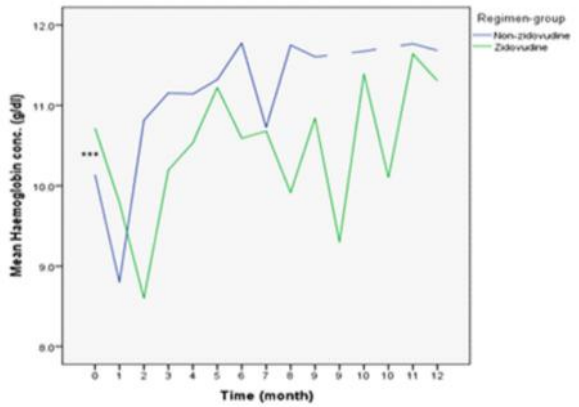

Fig. 2: Changes in haemoglobin concentration between the two regimen cohorts

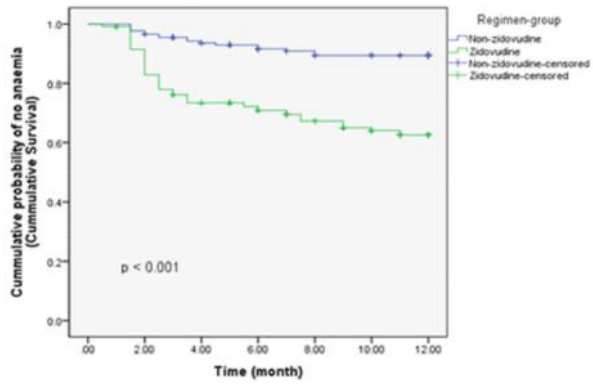

Fig. 3: Survival analysis of first occurrence of anaemia in patients without baseline anaemia by AZT treatment group

Incidence of anemia by treatment group

Patients initiated on AZT-containing ART (Group A) were 2.7 times more likely to develop anaemia than those receiving non-AZT containing ART during the first six month of initiation (p = 0.005) (Table 2a). Overall, the incidence of anaemia in patients initiated on an AZT-containing regimen was 32.4% (60.5 per 100 person-years) which is 4.52 times the incidence of anaemia among those receiving non-AZT containing ART regimen (P< 0.001; 95% CI: 2.55, 8.01). Among those with baseline anaemia (Group B), patients receiving AZT were 2.17 times more likely to develop worsening anaemia over baseline compared to those on non-AZT-containing regimens in the first six month of ART initiation (p = 0.013) (Table 2b). Overall, the incidence of worsening anaemia in AZT-containing regimens was 30.5% (57.5 per 100 person-years) which is significantly higher than the incidence of worsening anaemia in the non-AZT cohort (p < 0.001; OR: 3.11; 95% CI: 1.87, 5.17).

Figure 2 shows changes in hemoglobin across 12-months follow-up for both cohorts. Among patients receiving AZT, despite commencing ART at significantly higher haemoglobin concentrations (p = < 0.001), produced comparable haemoglobin concentration with non-AZT cohorts at end of the study. While the AZT cohort (both subgroups A2 and B2) curve produced several dips and was never associated with a peak higher than its baseline haemoglobin concentration, the non AZT was only associated with one distinct dip in the early stage of the study but characterized with several peaks above baseline hemoglobin concentrations during the length of the study.

Table 2(a): Incidence of anaemia in patients initiating zidovudine (AZT) compared with non-AZT containing regimens at selected time after initiation – Group A: patients without baseline anaemia

Time (month) after ART	AZT cohorts n= 225			Non-AZT cohorts n= 177			95% CI		
	event f (n)	Patient months	Incidence (per 100 patient Yrs).	Anaemia event f (n)	Patient months	Incidence (per 100 patient Yrs).	P value	OR	95% CI
0-6	63 (123)	1032	73.3	14 (50)	953.5	17.6	0.005*	2.70	1.33, 5.00
0-12	73 (225)	1448.5	60.5	17 (177)	1671.5	8.5	<0.001*	4.52	2.55, 8.01

**significant at p value < 0.05*

Table 2(b): Incidence of worsening anaemia in patients initiating zidovudine (AZT) compared with non-AZT containing regimens at selected time after initiation – Group B: patients with baseline anaemia

Time (months) after ART	AZT cohorts n= 175			Non-AZT cohorts n= 223			P value	OR	95% CI
	Anaemia event f (n)	Patient months	Incidence/ per 100 patient years.	Anaemia event f (n)	Patient months	Incidence/ 100 patient years			
0-6	43 (93)	782.5	65.9	25 (88)	1146.5	26.2	0.013*	2.17	1.17, 4.02
0-12	54 (177)	1127	57.5	28 (223)	1880.5	17.9	<0.001*	3.11	1.87, 5.17

**significant at p value < 0.05*

Cumulative incidence of anemia

The cumulative incidence of anaemia over 12 months for both AZT and non-AZT cohorts without baseline anaemia were 62.6% and 89.4% respectively (Figure 3). These survival curves were however significantly different in both cohorts ($p < 0.001$). Subjects on non-AZT regimens are significantly more likely to survive without anaemia of all type at 12 months than those with AZT-containing regimens.

Figure 3 shows that the 12-month cumulative survival of worsening anaemia of all causes for AZT and non-AZT in patients with baseline anaemia were 63.0% and 86.4% respectively. These survival curves were however significantly different in both cohorts ($p < 0.001$).

Table 3: Grades of first occurrence of AZT- related anaemia

WHO Grades	Hb Range	Anaemia f(% anaemia case)	%within sub-group[n=225]	Worsening Anaemia f(% anaemia case)	incidence within sub-group[n=175]	Total n=400 f(incidence rate within AZT cohort
1	9.5 - 10.5	12(30.00)	5.33	1 (2.04)	0.57	13 (3.25)
2	8 – 9.4	11(27.50)	4.89	20 (40.82)	11.43	31 (7.75)
3	6.5 - 7.9	10(25.00)	4.44	14 (28.57)	8.00	24 (6.00)
4	< 6.5	7 (17.50)	3.11	14 (28.57)	8.00	21 (5.25)
Total	=< 10.5	40 (100)	17.78	49 (100)	28.00	89 (22.25)

P value = 0.004, X^2 test

Table 4: Estimated time-to-event and survival time for AZT-related anaemia

Parameter	Event description (first occurrence)			Total
	Anaemia	Worsening anaemia	P value	
Median time-to-event in months (IQR))	2.25 (1.5, 3.38)	2.0 (1.5, 4.0)	0.750	2 (1.5, 3.75)
Average survival time in months (95% CI)	10.3 (9.8, 10.8)	9.2 (8.6, 9.9)	0.012*	9.8 (9.42, 10.2)

**significant at p = 0.05*

Table 5: Multivariate Analysis of factors associated with AZT-related Anaemia

Variable	Group	HR	95% CI for HR	P value
CD4+ < 200 (immunological AIDS)	No	Reference		
	Yes	NC	NC	.847
Baseline anaemia	No	Reference		
	Yes	.477	0.212 – 1.073	.074
Renal impairment	No	Reference		
	Yes	2.117	1.253 – 3.576	.005*
HCV infection	No	Reference		
	Yes	3.726	1.076 – 12.907	.038*
Thrombocytopenia	No	Reference		
	Yes	4.405	1.644 – 11.801	.003*
Cotrimoxazole use	No	Reference		
	Yes	9.014	3.615 – 22.478	< 0.001*
AIDS (CD4+ < 200 or WHO Stage 4 or both)	No	Reference		
	Yes	NC	NC	0.852
Baseline Hb (1-g/dl increment)	-	0.703	0.558 – 0.886	0.003*
Weight (1-kg increment)	-	0.982	0.952 – 1.012	0.241

**significant at p value < 0.05, NC: not computed*

Table 3 shows the categorization of the AZT-related anaemia using WHO toxicity grading and their respective incidence rate stratified by baseline anaemia status. Incidence of severe anaemia was 3.1% and 8.0% for the groups without baseline anaemia and with baseline anaemia, respectively. The group with baseline anaemia was more likely to develop severe anaemia (< 6.5 g/dl) (p = 0.004). Overall, the incidence of severe anaemia in AZT-cohorts was 5.3%. It contributed 23.6 % of all grades of anaemia in AZT-cohorts.

Time to new or worsening anemia

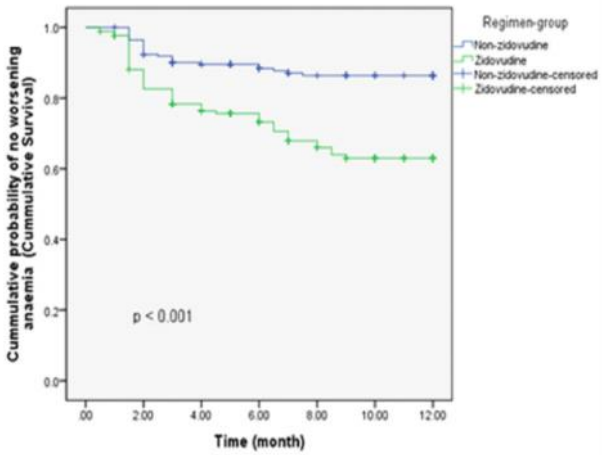

Fig. 4: Survival analysis of first worsening of anaemia in patients with baseline anaemia by AZT treatment group

Among patients without baseline anemia, the median time to first occurrence of anaemia was 2.3 months, versus 2.0 months for the first occurrence of worsening anaemia in the group with baseline anaemia, respectively (Table 4). The median time-to-event was however comparable in both groups whether commencing AZT-containing ART with baseline anaemia or not (p = 0.750). Overall, the median time-to-event was 2.0 months. The estimated 1-year median survival time was 10.3 months and 9.2 months for first occurrence of anaemia in group without baseline anaemia and for first worsening of anaemia in group with baseline anaemia, respectively. The estimated survival time was however longer in group without baseline anaemia than in group with baseline anaemia (p = 0.012). The overall estimated median survival time was 9.8 months.

The proportional hazards regression adjusted for confounding variables is as depicted in Table 5. The multivariate analysis indicates; renal impairment (p = 0.005, HR: 2.117), HCV infection (p = 0.038, HR: 3.726), thrombocytopenia (p = 0.003, HR: 4.405), concurrent cotrimoxazole use (p < 0.001, HR: 9.014) and baseline haemoglobin (p = 0.003, HR: 0.982) as independent predictors of AZT- related anaemia in patients on AZT-based HAART. While the renal impairment, HCV infection, thrombocytopenia, and concurrent cotrimoxazole use increase the incidence of AZT-related anaemia by 2.117, 3.726, 4.405 and 9.014 folds respectively, 1-g/dl increment in baseline haemoglobin independently decrease AZT- related anaemia by 2.8%.

DISCUSSION

Most data on HIV-related anaemia emanated from developed countries. However, the consequences of HIV-related anaemia is enormous and of great concern in resource-limited settings such as sub-Saharan Africa.[1,8,11-13] This current study evaluated the incidence of AZT-induced anemia among patients initiated on ART. Patients without anaemia initiated on ART including AZT were 2.7 times and 4.5 times likely to develop anaemia than patients initiated on non-AZT containing ART regimen when it was censored at 6 month and 12 month, respectively. The mean increase in haemoglobin concentration was significantly higher in cohorts initiated on non-AZT-containing ART regimen despite initiation on therapy with lower mean haemoglobin concentrations than those receiving AZT-containing ART. Several earlier studies have corroborated our findings of high incidence of anaemia among HIV-infected patients initiated[13,14] on AZT-containing ART regimens. Anaemia is a common feature of HIV-related disease and has been shown to independently predict morbidity and mortality irrespective of CD4 cell count and HIV viral load.[10,14] Although anaemia and other haematological abnormalities associated with HIV infection often respond to combination antiretroviral therapy, zidovudine (AZT), a commonly used component of antiretroviral therapy in resource-limited settings, is associated with myelotoxicity that often manifest as anaemia.[13,14] The AZT is associated with risk of myelosuppression that often manifest as anaemia than neutropenia and thrombocytopenia. It therefore increases the incidence of anaemia in patients on AZT oriented ART.[8,10,11,14]

The estimated median time to development of new or worsening AZT-related anaemia in this 12-month follow-up study, regardless of baseline anaemia, was 2.0 months (IQR: 1.5, 3.8). Further, when stratified by baseline anaemia status, the median time-to-event was comparable between the group with baseline anaemia and that without baseline anaemia. This median time-to-event was comparable to the findings of Ssali *et al.*[8] in Uganda; Curkendall *et al.*[14] in United States of America (USA); Katjitae *et al.* [15] in Namibia and Daka, Lelissa and Amsalu in Southern Ethiopia[12] and as well as a multi-center study from sub-Saharan Africa, Asia –Pacific and central and Southern America by Zhou *et al.*[16] The studies identified AZT-related anaemia as an early toxicity occurring mostly within the first six months. The finding of early AZT related toxicity is in agreement with the WHO recommendation on strict and close interval monitoring for anaemia in the first three months of AZT-containing ART.[17] However, despite WHO recommended guideline for monitoring of AZT-related haematological toxicity, reports from developing countries indicates poor adherence to the WHO recommended guidelines.[18,19] This lack of appropriate monitoring for early findings of reversible AZT-related anaemia may complicate treatment and worsen prognoses and subsequently undermine HIV intervention programs.[18-20]

The overall incidence of 22.3% (38.2 per 100 patient-years) obtained in our study was higher than previous studies despite using a lower cut-off value to define anaemia (haemoglobin <10.5 gm/dl), irrespective of gender. It was however similar to findings by Moh *et al.*[20] in Ivory Coast. Study design, cut off value for defining anaemia, and geographical location may all influence the anaemia incidence results.

In this report when AZT-related anaemia was censored at 6 months and further categorized using WHO toxicity grading, severe anaemia (haemoglobin < 6 g/dl) was reported in 5.3% irrespective of baseline anaemia and was significantly higher in patients with baseline anaemia. This finding is consistent with WHO estimates for adults and adolescents[17,21] and previous reports[22-24] which report an incidence of severe AZT-related anaemia in low and medium income countries of around 7%. Severe anaemia has been associated with poorer prognosis as it increases morbidity and mortality in HIV-infected patients.[25,26]

On multivariate analysis, HCV sero-positivity, renal impairment (ClCr <60ml/min) and concomitant cotrimoxazole were significantly associated with risk of AZT-related anaemia. AZT is metabolized by glucuronidation and its major metabolite is excreted through the renal system, thus in patients with renal impairment, failure to clear AZT may result in systemic accumulation of the drug, predisposing patients to AZT-related toxicity.[17,25]

Baseline Haemoglobin concentration correlated negatively with occurrence of AZT-related anaemia in the multivariate analysis, meaning that for every 1-g/dl increase in baseline haemoglobin the hazard for AZT-related anaemia was decreased by 29.7%. This finding is consistent with that of Curkendall *et al.*[14], who found 50.1% decreases in anaemia incidence for every 1-g/dl mean increase in baseline Haemoglobin, Katjitae *et al.*,[15] who found 12% increase in severe anaemia with every 1-g/dl decrease in baseline Haemoglobin. Both the WHO evidence based risk management consideration for prescription of AZT in the management of HIV infecion[17] and DART trial in Uganda[8] also identified significant correlation between baseline haemoglobin and subsequent risk of AZT-related anaemia.

Concomitant cotrimoxazole use was identified as a correlate by multivariate analysis for AZT- related anaemia, which implies that patients who had cotrimoxazole concomitantly with AZT-based ART are at risk of anaemia than those AZT cohorts without cotrimoxazole use. This observation is not consistent with report by Curkendall *et al.*[17] whose univariate proportional hazards analysis show association but failed significant test when it was adjusted for confounding variables in multivariate proportional hazards regression analysis. Randomized control trial in Uganda and Zimbabwe by Ssali *et al.*[8] also failed to demonstrate any correlation between anaemia incidence in ART and cotrimoxazole use. This association of cotrimoxazole use with AZT-related anaemia may be explained by the fact that cotrimoxazole, apart from its myelosppressive effect, has been reported to also contribute to zidovudine-induced anaemia by reducing the clearance of zidovudine through the kidney with resultant increase in the plasma level and subsequent toxicity.[27]

CONCLUSION

HIV-infected patients initiated on AZT-containing ART are 2.7 and 4.5 more likely to develop anaemia at 6 and 12 months respectively, than patients initiated on non-AZT containing regimens. When censored at 12 months the overall incidence of AZT-related anaemia was estimated to be 22.3% (38.2 per 100 person-years). The majority (75%) of AZT-related anaemia occurred early with estimated time-to-event occurring within the first 3.8 months (range, 0-15 weeks).

Recommendations

We recommend strict adherence of WHO guidelines of regular haemoglobin monitoring in patients initiating AZT-containing ART at week 4, 8, 14, and 24 to identify AZT-related anaemia as early as possible to avoid complication since it is an early medication toxicity.

REFERENCE

1. **Russell EC,** Charalambous S, Pemba L, *et al.* Low haemoglobin predicts early mortality among adults starting antiretroviral therapy in an HIV care programme in South Africa: a cohort study. BMC Public Health, 2010;10: 433
2. **Belperio PS,** Rhew DC. Prevalence and outcomes of anaemia in individuals with human immunodeficiency virus: a systemic review of literature. Am J Med. 2004;116 Suppl 7A: 27S-43S.
3. World Health Organisation (WHO), 2016. Antiretroviral medicines in low- and middle-income countries: forecasts of global and regional demand for 2015–2020, Geneva. (http://www.who.int/hiv/pub/amds/2013forecast_report/en, accessed 28 March 2018).
4. **Moyle G,** Sawyer W, Law M, *et al.* Changes in hematologic parameters and efficacy of thymidine analogue-based, highly active antiretroviral therapy: a meta-analysis of six prospective, randomized, comparative studies. Clinical Therapeutics 2004; 26 (1): 92-97.
5. **Moore RD,** Forney D. Anemia in HIV-infected patients receiving highly active antiretroviral therapy. J Acquir Immune Defic Syndr 2002; 29 (1): 54–57.
6. **Sharma SK.** Zidovudine-induced anaemia in HIV/AIDS. Indian J Med Res 2010; 132 (10): 359-361.
7. **Weinkove R,** Rangarajan S., van der Walt J Kulasegaram, R. Zidovudine-induced pure red cell aplasia presenting after 4 years of therapy. *AIDS*. 2005; 19 (17): 2046-2047.
8. **Ssali F.,** Stöhr W., Munderi P., *et al.* Prevalence, incidence and predictors of severe anaemia with zidovudine-containing regimens in African adults with HIV infection within the DART trial. Antivir Ther.2006; 11(6):741-749
9. **Ibrahim, T (2009).** Research methodology and dissertation writing for health and allied health professionals. Abuja, Nigeria. Cress global link limited.
10. **Sullivan PS,** Hanson DL., Chu SY., *et al.* Epidemiology of anemia in human immunodeficiency virus (HIV)-infected persons: results from the Multistate Adult and Adolescent Spectrum of HIV Disease Surveillance Project. The Adult/Adolescent Spectrum of Disease Group. Blood. 1998; 91(1):301–308
11. **Agarwal D,** Chakratarty J, Chaube L, *et al.* High incidence of zidovudine induced anaemia in HIV infected patients in eastern India. India J Med Res 2010;132(10):386-389.
12. **Daka D,** Lelissa D, Amsalu A. Prevalence of anaemia before and after the initiation of antiretroviral therapy at ART centre of Hawassa University Referral Hospital, Hawassa, South Ethiopia. Sch J Med. 2013;3(1):1-6.
13. **Kuwalairat P,** Wint-Watjana W, Chumphon Community Hospital Antiretroviral Clinic Group. Determinant for zidovudine induced in HIV adult patients: A Thai Multicenter study.Arch Pharma Pract 2014; 5 (1):6-13.
14. **Curkendall SM,** Richardson JT, Emons MF, *et al.* Incidence of anaemia among HIV-infected patients treated with highly active antiretroviral therapy. British HIV Association HIV Medicine 2007; 8(8):483–490

15. **Katjitae I,** Mengistu A, Corbell C, *et al.* Risk of anaemia associated with zidovudine (AZT)-based HAART in Namibia 2010; 9:236-446.
16. **Zhou J,** Jaquet A, Bissagnene E, *et al.* Short-term risk of anaemia following initiation of combination antiretroviral treatment in HIV-infected patients in countries in sub-Saharan Africa, Asian – Pacific and central and South America. J int AIDS Soc 2012;15:5.doi:10.1186/1758-2658-15-5.
17. World Health Organisation. (2010). WHO: Antiretroviral therapy for HIV infection in adults and adolescents, Recommendations for a public health approach. Available at: http://whqlibdoc.who.int/publications/2010/9789241599764_eng.pdf accessed on 12 October, 2014
18. **Egger M,** Ledergerber B, Lundgren, J. Prognostic importance of Anaemia in HIV type-1- infected patients starting antiretroviral therapy: Collaborative analysis of prospective cohort studies. International medical press 1359-6535, antiviral therapy: 2008;13(8): 959-967
19. **Volberding PA,** Levine AM, Dieterich D, *et al.* Anaemia in HIV infection: clinical impact and evidence-based management strategies. Clin Infect Dis 2004;38 (10):1454 – 1463.
20. **Moh R,** Danel C, Sorho S, *et al.* Haematological changes in adults receiving a zidovudine – containing HAART regimen in combination with cotrimaxazole in Cote d Ivoire. Antiviral Therapy 2005;10:615 -624.
21. World Health Organization. Scaling Up Antiretroviral Therapy in Resource Limited Settings: Treatment Guidelines for a Public Health Approach - 2003. Revision available at: http://www.whoint/3by5/publications/guidelines/en/execsumm.pdf, accessed December 2016.
22. **Phe T,** Thai S, Veng C, *et al.* Risk factors of treatment-limiting anaemia after substitution of zidovudine for stavudine in HIV-infected adult patients on antiretroviral treatment. PLoS One 2013; 8(3): e60206.
23. **Eluwa GI,** Badru T, Agu KA, *et al.* Adverse drug reaction to antiretroviral therapy (ARVs): incidence, type and risk factors in Nigeria. BMC Clin Pharmacol 2012;12: 7.doi.1186/1472-6904-12-7.
24. **Max B,** Sherer R. Management of the adverse effects of antiretroviral therapy and medical adherence. Clinical Infectious Diseases 2000;30 (suppl 2): S96 – 116.
25. **Murphy RA,** Sunpath H, Kuritzkes DR, *et al.* Antiretroviral Therapy–Associated Toxicities in the Resource-Poor World: The Challenge of a Limited Formulary.JID 2007;196 (Suppl 3): S452. Available at: http://jid.oxfordjournals.org/[accessed on 20th July 2014]
26. **Ogoina D,** Obiako RO, Muktar HM *et al.* Morbidity and mortality patterns of hospitalised adult HIV/AIDS patients in the era of highly active antiretroviral therapy: A 4-year retrospective review from Zaria, Northern Nigeria. Hindawi Publishing Corporation; AIDS Research and Treatment Volume 2012, Article ID 940580: 2-9
27. FMOH. National guidelines for HIV/AIDS Treatment and Care in adolescents and adults In: National AIDS/STIs Control Programme, editor. Abuja: FMOH; 2010. p. 12–13. [Online] Available at: http://www.who.int/hiv/pub/guidelines/nigeria_art.pdf [Accessed on 29th September 2014]