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Risk of zidovudine-induced anemia on human immunodeficiency virus (HIV) infection patients with different CD_4 cell counts

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ABSTRACT

Anemia is the most common hematologic abnormality in patients with human immunodeficiency virus (HIV) infection. This abnormality is associated with HIV infection itself, HIV-related opportunities infections or drug use. Zidovudine (AZT) is the most common cause of anemia in HIV patients. Recent study showed anemia in HIV patients is also associated with CD_4 cell counts. Aim of this study was to evaluate the risk of anemia on HIV patients with different CD_4 cell counts after AZT-based antiretroviral therapy (ART).This retrospective cohort study was conducted using medical record of HIV patients in Dr. Soetomo General Hospital, Surabaya. Subjects who fulfilled the inclusion and exclusion criteria were divided into two group i.e. HIV patients with CD_4 cell counts 200-350 cell/mm^3 and those with CD_4cell counts $\geq$350 cell/mm^3. All available demographics, clinical and laboratory data of subjects before and after AZT-based ART were then recorded and evaluated. Ninety-seven HIV patients (50 male and 47 female) were involved in this study. The result showed that the anemia incidence significantly increased after AZT-based ART ($p<0.05$), however no significantly different in anemia incidence, mean Hb level reduction and Hb level time reduction were observed between HIV patients with CD_4 cell counts 200-350 cell/mm^3 and those with CD_4cell counts $\geq$350 cell/mm^3($p>0.05$). Gender, age, weight and clinical stage were not associated with anemia incidence ($p>0.05$). In contrast, anemia incidence is associated with Hb level before AZT therapy ($p<0.05$). In conclusion, the anemia incidence in HIV patients after AZT based ART is not associated with the level of CD_4 cell counts, however it is associated with Hb levels before AZT therapy.

ABSTRAK

Anemia adalah kelainan hematologi yang paling sering dijumpai pada pasien yang terinfeksi *human immunodeficiency virus* (HIV). Kelainan ini dikaitkan dengan infeksi HIV itu sendiri, infeksi oportunitis yang berkaitan dengan HIV dan penggunaan obat. Zidovudin

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(AZT) adalah penyebab anemia tersering pada pasien HIV. Penelitian terkini menunjukkan anemia pada penderita HIV juga dikaitkan dengan kadar CD_4 pasien. Penelitian ini bertujuan mengkaji risiko anemia pada pasien HIV dengan kadar CD_4 yang berbeda setelah menerima pengobatan anti retrovirus berbasis AZT. Penelitian *cohort retrospective* ini menggunakan data rekam medik pasien HIV di RSUP Dr. Soetomo, Surabaya. Subjek yang memenuhi kriteria inklusi dan eksklusi dibagi menjadi dua kelompok yaitu pasien HIV dengan kadar CD_4 200-350 sel/mm^3 dan pasien dengan kadar $CD_4 > 350$ sel/mm^3. Semua data demografi, klinik dan laboratorium subjek yang ada sebelum dan sesudah pengobatan anti retrovirus berbasis AZT dicatat dan dievaluasi. Sebanyak 97 pasien terdiri dari 50 laki-laki dan 47 wanita diambil datanya dalam penelitian ini. Hasil penelitian menunjukkan kejadian anemia meningkat secara nyata setelah pengobatan AZT pada kedua kelompok ($p < 0,05$). Namun demikian, kejadian anemia, rerata penurunan kadar Hb, dan waktu penurunan kadar Hb tidak berbeda nyata pada pasien dengan kadar CD_4 200-350 sel/mm^3 dan pasien dengan kadar $CD_4 > 350$ sel/mm^3 ($p > 0,05$). Jenis kelamin, umur dan berat badan tidak berkaitan dengan kejadian anemia ($p > 0,05$), akan tetapi kadar Hb sebelum pengobatan dengan AZT berkaitan nyata dengan kejadian anemia ($p < 0,05$). Dapat disimpulkan, kejadian anemia pada pasien HIV setelah pengobatan anti retrovirus berbasis AZT tidak berkaitan dengan kadar CD_4 tetapi berkaitan dengan kadar Hb sebelum terapai AZT.

Key words: human immunodeficiency virus (HIV), anemia, zidovudine, CD_4 cell counts, adverse drug event

INTRODUCTION

Anemia is the most common haematologic abnormality in patients with human immunodeficiency virus (HIV) infection. Anemia affects 18% of patients with asymptomatic HIV-seropositive, 50% of those with middle-stage HIV disease and 75% of those with late-stage HIV disease.[1,2] An obvious cause of anemia in patients with HIV infection is blood loss. Other than blood loss, the pathophysiology of HIV associated anemia may involve decreased red blood cell (RBC) production, increased RBC destruction and ineffective RBC production.[1,3] Factors associated with anemia in HIV patients include HIV infection itself, HIV-related opportunities infections or as a consequence of drug used for HIV infection and associated condition.[4,5]

Several drugs that commonly cause anemia in HIV patients have been reported.[1,4,5] Among these drugs, zidovudine (AZT) is probably the most common cause of anemia in HIV patients. Zidovudine, a nucleoside reverse transcriptase inhibitor (NRTI), is one of earliest antiretroviral drugs used as a combination in some of the highly active antiretroviral therapy (HAART) regimens to treat of HIV/AIDS. Moreover, AZT is the first drug which was approved by US FDA for use in HIV/AIDS.[6] Its used, however, is associated with hematologic abnormalities especially anemia. This anemia is observed in most of the patients within 3-6 months of initiation of AZT therapy. Female patient has been found to be a risk factor for anemia in some studies.[6-8]

Another factor currently associated with anemia in HIV patients is CD_4 cell counts. It was reported that 3.2% of asymptomatic HIV-seropositive patients with CD_4 cell counts greater than 700 cells/mm^3 were anemic, where as anemia was observed in 20.9% of those with CD_4 cell counts less than 249 cells/mm^3.[9] Furthermore, CD_4 cell counts less than

200 cells/mm^3 was reported as a risk factor associated with anemia in HIV patients. Anemia was observed in 91.4% of HIV-seropositive patients with CD4 cell counts less than 200 cells/mm^3. [8,10] This study was conducted to evaluate therisk of anemia in human immunodeficiency virus (HIV) infection patients with different CD_4 cell counts after AZT therapy.

MATERIALS AND METHODS

Research design and subjects

This retrospective cohort study was conducted using data from medical record of HIV patients who attended at Dr. Soetomo General Hospital, Surabaya between January 2005 to August 2015. The subjects were selected with the inclusion criteria as follows HIV patients underwent AZT-based therapy, aged 21-50 years, having CD_4 cell counts ≥ 200 cell/mm^3 before AZT-based antiretrovirus therapy (ART), having hemoglobin (Hb) level ≥ 10 and body mass index (BMI) 19-25. The exclusion criteria were pregnant women, underwent cotrimoxazole therapy, suffered from tuberculosis and received oral anti tuberculosis drugs, suffered certain diseases causing anemia such as renal chronic diseases and worm infection. Written inform consent was obtained from patients. The protocol of the study was approved by the Medical and Health Research Ethics Committee, Faculty of Medicine, Universitas Gadjah Mada, Yogyakarta.

Procedure

Subjects who met the inclusion and exclusion criteria were divided into two groups. The first group was HIV patients with CD_4 cell counts 200-350 cell/mm^3, and the second group was HIV patients with CD_4 cell counts ≥350 cell/mm^3. All available demographics (age, gender, body height and body weight), clinical (history of diseases and history of treatment) and laboratory (viral load counts, CD_4 cell counts and Hb level) data of subjects before AZT-based ART were then recorded. After AZT-based ART for six months, clinical and laboratory data of both groups were monitored and recorded again. A number of additional factors that considered as potential confounders were recorded such as opportunistic infections, renal insufficiency were recorded and analyzed.

Data analysis

All recorded demographics, clinical and laboratory data were presented as mean ± standard deviation (SD) or number and percentage. Univariate analysis was applied to evaluate the demographics, clinical and laboratory data. Bivariate analysis such as chi-square ort-test was applied to evaluate the mean Hb level reduction and Hb level time reduction between group. Multivariate analysis using analysis of variance (ANOVA) was applied to evaluate the relationship between confounding factors and dependent variable.p value less than 0.05 was considered statistically significant.

RESULTS

During the study period 97 HIV patients who underwent AZT based ART were included in this study. Of these included patients, 50 (51.5%) were male and 47 (48.5%) female. The age of patients ranged from 26 to 46 years old. Most of these included patients had body weight ranged from 40 to 60 kg and in stage III of HIV infection. Furthermore, most of these included patients suffered from HIV infection through heterosexual transmission.

TABLE 1. HIV patient characteristics who underwent AZT based ART in Dr. Soetomo General Hospital, Surabaya between January 2008 and August 2015.

Characteristics	n (%)
Gender	
• Man	50(51.5)
• Woman	47(48.5)
Age (years)	
• > 46	18(18.6)
• 46-26	68(70.1)
• < 26	11(11.3)
Education	
• Elementary School	11(11.3)
• Secondary School	23(23.7)
• Senior High School	46(47.4)
• University	17(17.5)
Occupation	
• Employees	65 (67.0)
• Daily labor	18(18.6)
• Permanent employment	23(23.7)
• Private employment	1(1.0)
• Unemployment	33 (33.0)
Clinical stage	
• I	22 (22.7)
• II	16 (16.5)
• III	42 (43.3)
• IV	17 (17.5)
Body Weights (kg)	
• <40	7 (7.2)
• 40 – 60	71 (7.2)
• >60	19 (9.6)
Transmission	
• Heterosexual	51(52.6)
• Homosexual	18(18.6)
• Heterosexual + homosexual	18(18.6)
• Drug ubuse	1(1.0)
• Transfusion	2(2.1)
• Heterosexual, + drug ubuse	6(6.2)
• Unknown	2(2.1)

This result showed that the anemia incidence significantly increased after AZT based ART comparae to beforethose both in HIV patients with CD_4 cell counts 200-350 cell/$mm^3$and those ≥350 cell/mm^3 (TABLE 2). However, it was not affected by the level of CD_4 cell counts.The anemia incidence on patients with CD_4 cell counts 200-350 cell/mm^3 was not significantly different compared to those with CD_4 cell counts >350 cell/mm^3 (TABLE 3).

TABLE 2. Hemoglobin level of HIV patients before and after AZT based ART in CD4 cell counts 200-350 cell/mm3and >350 cell/mm3 in Dr. Soetomo General Hospital, Surabaya between January 2005 and August 2008

Hb level	Before AZT	After AZT	Total	p
$CD_4$200-350				
• Severe	5	13	18	
• Moderate	2	23	25	
• Mild	5	11	16	<0.05
• Very mild	17	14	31	
• Normal	32	0	32	
CD4> 350				
• Severe	0	6	6	
• Moderate	4	12	16	
• Mild	4	9	13	< 0.001
• Very mild	7	9	16	
• Normal	21	0	21	

chi-square with $p<0.05$

The CD_4 cell counts, gender, age, body weight and clinical stage were not associated with the anemia incidence in HIV patients after AZT based ART (TABLE 4). However, the anemia incidence in those patients was significantly associated with the level of Hb before AZT based ART ($p<0.05$).

TABLE 3. Hemoglobin level of HIV patients with different level of CD4 cell counts before and after AZT based ART in Dr. Soetomo General Hospital, Surabaya between January 2005 and August 2008

Hb level	CD_4 cell counts (cell/mm^3)		RR 95% CI	p
	200 - 350 n (%)	> 350 n (%)		
Hb before AZT				
• Severe	5	0	3.94 (0.41-93.15)	0.185
• Moderate	2	4	0.33 (0.04-2.38)	
• Mild	5	4	0.82 (0.016-4.21)	
• Very mild	17	7	0.59 (0.51-5.13)	
• Normal	32	21	1	
Hb after AZT				
• Severe	13	6	1.39 (0.32-6.08)	0.815
• Moderate	23	12	1.23 (0.36-4.21)	
• Mild	11	9	0.79 (0.20-3.14)	
• Very mild	14	9	1	

chi-square with $p<0.05$

TABLE 4. The relationship of patients' predictors to anemia incidence in HIV patients after AZT based ART in Dr. Soetomo General Hospital, Surabaya between January 2008 and August 2015

Variable	Hb level*		RR (95% CI)	p**
	Anemia n (%)	Normal n (%)		
CD4 (sel/mm^3)				
• 200 – 350	43 (70.5%)	18 (29.5%)	0.919 (0.369-2.291)	0.523
• > 350	26 (72.2%)	10 (27.8%)	1	
Gender				
• Man	35 (70.0%)	15 (30.0%)	0.892 (0.370-2.151)	0.488
• Woman	34 (72.3%)	13 (27.7%)	1	
Age (years)				
• > 46	15 (83.3%)	3 (16.7%)	1.88 (0.22-16.14)	
• 46-26	46 (67.6%)	22 (32.4%)	0.78 (0.15-3.74)	0.423
• < 26	8 (72.7%)	3 (27.3%)	1	
Weight (kg)				
• < 40	6 (85.7%)	1 (14.3%)	1.6 (0.11-46.01)	
• 40-60	48 (67.6%)	23 (32.4%)	0.56 (0.14-2.08)	0.423
• > 60	15 (78.9%)	4 (21.1%)	1	
Clinical stage				
• 3 – 4	43 (72.9%)	16 (27.1%)	1.240 (0.508-3.029)	0.401
• 1 – 2	26 (68.4%)	12 (31.6%)	1	
Hb before AZT (g/dL)				
• Anemia	45 (80.4%)	11 (19.6%)	2.898 (1.171-7.169)	0.017
• Normal	24 (58.5%)	17 (41.5%)	1	

* anemia if Hb level < 10 g/dL and normal if >10 g/dL; **chi-square with $p<0.05$

No significant difference in mean reduction and time reduction of Hb level between HIV patients with CD_4 cell counts of 200- 350 cells/$mm^3$and those with CD_4 cell counts >350 cells/mm^3were observed in this study (TABLE 5 and 6).

TABLE 5. Mean Hb level reduction (mean ± SD) before and after AZT based ART of HIV patients with different CD4 cell counts in Dr. Soetomo General Hospital between January 2008 and August 2015

Hb's level	CD_4 cell counts		Mean diff (95% CI)	p
	200-350(n=61)	>350(n=36)		
Before	11.34 ± 1.82	11.67 ±1.69	-0.322 (-1.061-0.416)	0.388
After	8.14 ± 2.16	8.41 ± 2.25	-0.265 (-1.180-0.651)	0.568
Mean reduction	3.20 ± 2.11	3.26 ± 2.24	-0.058 (-1.180-0.651)	0.899

t-test with $p<0.05$

TABLE 6. Time reduction of Hb level (days) after AZT based in HIV patients in Dr. Soetomo General Hospital, Surabaya between January 2008 and August 2015

CD_4 cell counts		Mean diff (95% CI)	p
200-350(n=61)	>350(n=36)		
34.03±23.01	37±21.27	-3.467(-12.809-5.874)	0.463

t-test with $p<0.05$

DISCUSSION

This study showed that the anemia incidence significantly increased after AZT based ART in HIV patients (TABLE 2).It is ndicated that AZT can induce anemia in HIV patients. Zidovudine treatment is associated with bone marrow suppression and an increased risk of developing anemia.[1]This result is suported with the previous studies reported by some authors. Women's Interagency HIV Study (WIHS) reported the presence of anemia in 42.6% of HIV-infected women patients receiving AZT therapy compared with 34.3% of those not receiving AZT.[3] Furthermore, Agarwal *et al*.[11] reported a high incidence of AZT-induced anemia (42.7%) in HIV-infected patients from eastern part of India. In contrast Semba *et al.*[12] reported that, although the use of zidovudine was associated with an increased risk of anemia in the pre-HAART era (1993–1996), use of zidovudine during the HAART era (1996–2000) was not significantly associated with anemia.

The study also showed that the anemia incidence was not associated with the level of CD_4 cell counts. The anemia incidence on patients with CD_4 cell counts 200-350 cell/mm^3 was not significantly different compared to those with CD_4 cell counts >350 cell/mm^3 (TABLE 3). It indicated that anemia incidence in HIV patients is not affected by the level of CD_4 cell counts. The association between CD_4 cell counts and anemia incidince is not clear. Previous studies the decrease of CD_4 cell counts was independently associated with the increase of anemia risk.Anemia affected 30% of HIV patients with CD_4 cell counts ≥ 500 cell/mm^3 and 63-95% of those with

CD_4 cell counts < 200 cell/mm^3.[8] In addition, anemia more frequent affected HIV patients with CD_4 cell counts < 350 cell/mm^3.[13] In contrast, another study reported that there was no association between anemia incidence and CD_4 cell counts.[14]Similarly a study among pregnant HIV positive women reported that there was no significant association between Hb level and CD4 cell counts.[15]

Although gender, age, weight and clinical stage have reported as risk factor for anemia in HIV patients in previous studies, [1,3,4,16] in the present study did not find as risk factor. However, the level of Hb before based ART has been found as risk factor in this study. The different in study designs, methodologies use including inclusion and exclusion criteria and cut-offs used to define anemia may contribute to these defferences.

CONCLUSION

In conclusion, there are no differences in the risk of anemia, a mean reduction of Hb level and time reduction of Hb level between HIV patients after AZT based ART with CD_4 cell counts 200-350 cell/mm^3 and those CD_4 cell counts >350 cell/mm^3. The anemia incidence in HIV patients after AZT based ART is associated with the Hb levels before AZT therapy.

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