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Factors Affecting CD4 Count Response in HIV Patients within 12 Months of Treatment: A Case Study of Tamale Teaching Hospital

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Abstract The effectiveness of HIV treatment can be assessed by monitoring the Cluster of Differentiation 4 (CD4) cell counts of HIV positive patients. Changes in CD4 cell counts over time in patients on antiretroviral therapy (ART) could explain how HIV patients respond to treatment (ART). We seek to identify factors that affect Cluster of Differentiation 4 (CD4) cell count response in patients on antiretroviral therapy (ART) within 12 months of treatment at the Tamale Teaching Hospital (TTH) of Ghana. The data was based on the records of patients in the database of the hospital from 2009 to 2013. Factors identified include social (smoking habits, religious affiliation and alcohol consumption) and demographic factors (age, sex and employment status); antiretroviral therapy (ART regimen) and immunological CD4 cell count at the initiation of treatment). However, only two of the variables (Age and Gender) were found to be significant and were therefore included in the model. The rest of the variables did not affect CD4 cell count response to antiretroviral therapy (ART). The results also showed that the model provided a reasonable statistical fit (chi-square value of 10.058 with corresponding probability value of 0.261 which is greater than the level of significance (0.05)). Since age affected (increase) in the CD4 cell counts in response to ART, detection of HIV infections at young age is paramount for early treatment when there the likelihood of significant CD4 cell count increase. This will translate into increased immunity and prolong the lives of HIV patients.

Keywords: *CD4 count, HIV, AIDS, Logistic Regression, antiretroviral therapy*

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1. Introduction

When a person is infected with HIV, the virus enters the body and then multiplies primarily in the white blood cells (the immune cells which protect the body from diseases). Overtime, the virus damages or kills specific immune cells, weakening the immune system and leaving the body vulnerable for other infections. One is diagnosed with AIDS only when one begins to experience one or more of the conditions (associated with HIV and AIDS) such as loss of significant amount of immune cells, mild and persistent swollen glands. An individual may look and feel healthy, (even if infected); the only way to be certain about one's status is to test for HIV [1]. AIDS is linked with deficiency of glutathione resulting to generation of huge levels of oxidative stress which impair and kills healthy cells as well as decrease the immune system of the body [2]. About 36.7 million people globally were living with HIV as at 2015, with 17 million people accessing antiretroviral therapy. Since the start of the epidemic about 78 million people has been infected with HIV with 35 million people dying from AIDS and related illnesses [1]. Furthermore, young people (under 25 years) accounted for half of all the new HIV infections worldwide, 22 million adults and children were living with HIV and AIDS and an estimated 1.5 million Africans died from AIDS [1]. The epidemic was estimated to have orphaned about 11.6 million African children [3]. Globally, new HIV infection (cases) declined by 33% between 2005 and 2013 with Sub-Saharan Africa accounting for about 70% of the global infections [4]. There have been about 43% declines in new HIV infections among children in 21 priority countries of the Global Plan in Africa [4]. In Ghana, HIV prevalence among antenatal clients in 2015 was 1.8% a slight increase (1.6%) from in the 2014 cases. However, HIV prevalence among the young population (age 15-24) went down by 40% from 50% in 2014. This age group (age 15-24) constitutes a proxy for new infections. Hence, we must sustain interventions in this target group (age 15-24) to ensure sustainability of the gains [5]. An estimated 270, 000 people lived with HIV in Ghana as at December 2015, with 260, 000 been adults (aged 15 and over). Meanwhile, women (age 15 and over) accounted for about150, 000 (50%) of the adult cases, whereas about 10,000 children (age 0 to 14) lived with HIV. Within the same year (2015)

160, 000 (59%) children (age 0 to 17) were reported to be orphaned due to AIDS. Whiles the number of deaths due to AIDS was 13,000 (5%) in 2015 [6]. In the meantime, about 90,573 (adult and children) patients were introduced to Antiretroviral therapy (ART) in Ghana by the end of 2013; 5.6% of whom were children (under 15 years), 29.9% were males and 71% of the patients under ART were alive 12 months post ART initiation [7]. Cluster of Differentiation 4 (CD4) cell count is one of the most important laboratory indicators of immune status in HIV-infected patients [12]. It is the strongest predictor of subsequent disease progression and survival of HIV-infected patients [8]. T-lymphocytes helper cells (containing CD4+ receptor) are usually infected by HIV-1 on the surface macrophages or dendridic cells and destroy them rapidly resulting to sharp decline in their (T-lymphocytes) counts. Consequently, complete collapse of cell- mediated immunity occurs (when the number of CD4+ T- lymphocytes declined below a critical level 200/ul) and the body becomes prone to opportunistic infections (by pathogens). Entry of HIV-1 into CD4+ T cells (or macrophages) occurs via interaction between the glycoproteins (gp120) on the viral envelope and the CD4 receptor on targeted cells [9]. As such particular co-receptors such as chemokine receptor type 4 (CXCR-4) for T-lymphocytes or CC chemokine receptor 5 (CCCR5) for macrophages are needed for the internalization of virus into the cells after docking [9]. Even so, HIV patients are encouraged to continue taking medication to reduce the viral load and to increase the blood count of CD4+ T-lymphocytes cells [10]. When this regimens(treatments) is observed routinely, it arrest noticeable side effects by developing adverse reactions, leading to many metabolic disorders in HIV patients and, exerts a negative impact on adherence [10]. Moreover, the frequency of antiretroviral (ARVs) failures due to recurrent viral mutations (as well as toxicity) in HIV patients lead to failure of chemotherapy against AIDS [11]. Beside, appropriate retroviral drug development and applications, other approaches such as increase ARVs adherence, drug resistance genotyping, regular monitoring and surveillance of drug resistance, varying retroviral (to resistance) and selecting best drugs combinations may be employed to combat HIV successfully [12]. The World Health Organization (WHO) currently recommends that HIV-positive adults start ART to CD4 cell counts of less than 350cells/mm^3.However, several countries have changed their guidelines on ART at CD4 cell count(threshold) of 500 cells/ mm^3 [13]. There are two major parts of the immune system; one (antibody mediated) which works through antibodies (produced by B- cells and plasma cells) and another (cell mediated) which works through other cells including CD4 Lymphocyte cells. The cell mediated immune system is suppressed in people diagnosed with AIDS [14]. In fact, majority of the HIV patients have less than 200cells/mm^3 CD4 cell counts due to late start of ART treatments [15]. With effective ART, CD4 cell count may increase up to more than 50 cells /mm^3 within weeks after viral suppression [16]. For this reason ART services have been available to HIV patients in Ghana since 2001 [17]. The Ghana National AIDS Control Programme adopted the WHO recommendation that HIV patients be initiated into antiretroviral drugs (treatment) when their CD4 count is less than 350 cells mm^3. HIV prevalence in Ghana varies with geography, gender, age and residence [18]. It has been revealed that age is a significant predictor of CD4 count as well as overall health status of an HIV patient; with older patients at higher risk [19]. Islam as a religion contributed to decrease in the average CD4 count of patients (after the first 6 months of treatment) whereas the levels of education and treatment regimen and religious affiliations are significantly different at 5 % significance level (P-Values < 0.05) [20]. Age at the start of ART, educational level, marital status and baseline of CD4 count are also predictors of CD4 count. [21]. Yet there was no evidence that alcohol consumption affected changes in CD4 cell count in [22]. However, after CD4 cell count baseline correction; viral load and use of antiretroviral drugs, the frequency of alcohol use almost tripled the risk of drop in CD4 cell count [23]. Educational level and marital status are also seen as significant predictors of CD4 cell count in HIV patients [24]. In this study we seek to identify factors that affect CD4 count response in patients on Antiretroviral Therapy within 12 months of treatment at the Tamale Teaching Hospital of Ghana using logistic regression model and SPSS version 20.0 to analyze the data.

2. Materials and Methodology

The medical records (300 files) of HIV and AIDS patients in the database of the Tamale Teaching Hospital (T.T.H) from 2009 to 2013 were used to extract the data for the qualitative analysis. All patients were 14 years and above and had their initial CD4 cells counted at least twice (within the first 12 months of initiation into ART). The CD4 cell count on four different drug combinations including (Zidovudine, Lamivudine and Nevirapine; Tenofovir, Lamivudine and Nevirapine; Tenofovir, Lamivudine and Efavirenz and finally, Zidovudine, Lamivudine and Efavirenz were considered and the database (Microsoft access) exported to SPSS for the subsequent analysis.

2.1. Data Collection and Analysis

Changes in CD4 cell counts in every six months were also examined and stratified on CD4 cell count of ≤50-100 cell counts/ mm^3 baseline. Control data were those whose CD4 cell counts increased (100cells/mm^3) above the baseline (≥50-100 cells/mm^3) per year in response to 12 months of ART. Using deviance concept of with Wald Statistics, the study variables were subjected to statistical testing; categorical variables were summarized as percentages and frequencies whereas numerical variables with non-normal distribution were summarized as median and mean. Factors associated with risks of immunological non-response (failure to increase CD4 cell counts at 12 months of ART) were examined by logistic regression. Tests of significance were two-sided with (probability of less than $p<0.05$) indicating statistical significance.

2.2. Modeling

Logistic regression model was employed (as the statistical method) to analyze the data using SPSS for Windows (version 20.0) and to predict the relative likelihood of the effect of CD4 cell count on ART.

Furthermore, Wald tests, Odds Ratio, Hosmer and Lemeshow Test, Cox and Snell R-Square Test, (with Deviance) and Chi-square test were employed for the analysis. The dependent variable (CD4 cell counts) dichotomous as such SPSS version 20.0 software package was used for the analysis.

2.3. The Logistic Regression Model

Logistic regression is the usual regression analysis tool used when response variables are binary in nature. The model is used to predict the likelihood of the increase of CD4 cell counts because it (model) provides the best fitting of the relationship between a binary dependent variable and the independent (explanatory) variables. The dependent variable is the probability (p) that the outcome is equal to 1; for each of the independent variables in the model, the parameters obtained are used to estimate the odds ratios [1]. Moreover, the binary response variable (x) is categorized by success (1) and failure (0) as the outcomes based on the logistic transformation (logic of proportionality) as:

$$p(x)=\frac{\exp^{(\beta_0+\beta_1 x)}}{1+\exp^{(\beta_0+\beta_1 x)}} \quad (1)$$

$$1-p(x)=1-\frac{\exp^{(\beta_0+\beta_1 x)}}{1+\exp^{(\beta_0+\beta_1 x)}}1$$

$$=\frac{\left(1+\exp^{(\beta_0+\beta_1 x)}\right)-\exp^{(\beta_0+\beta_1 x)}}{1+\exp^{(\beta_0+\beta_1 x)}}$$

$$1-p(x)=\frac{1}{1+\exp^{(\beta_0+\beta_1 x)}}$$

Writing the logistic regression in terms of Odds result to

$$Odds=\left(\frac{p(x)}{1-p(x)}\right) \quad (2)$$

This can be obtained as;

$$\frac{p(x)}{1-p(x)}=\frac{\exp^{(\beta_0+\beta_1 x)}}{1+\exp^{(\beta_0+\beta_1 x)}}\div\frac{1}{1+\exp^{(\beta_0+\beta_1 x)}}$$

$$=\frac{\exp^{(\beta_0+\beta_1 x)}}{1+\exp^{(\beta_0+\beta_1 x)}}\times\frac{1+\exp^{(\beta_0+\beta_1 x)}}{1}$$

$$\frac{p(x)}{1-p(x)}=\exp^{(\beta_0+\beta_1 x)}$$

Logarithm of the odds leads to

$$g(x)=\log\left(\frac{p(x)}{1-p(x)}\right)=\beta_0+\beta_1 x \quad (3)$$

Transformation of logistic function (logit transformation) yields

$$\log\left(\frac{p(x)}{1-p(x)}\right)=\beta_0+\beta_1 x$$

Where p is the probability of the dependent variable (p =1), x is the explanatory variables B_0 is the coefficient of the constant (intercept) in the model, $\beta_1 x$ is the coefficient of a variable in the model; Log is Logarithm; exp. is the exponentiation of the B coefficient which is an odds ratio.

3. Results and Discussion

3.1. Data Presentation

Table 1 shows the descriptive statistics and the results of the logistic regression analysis. After a retrospective review of medical records, 300 files of clients who were registered for ART at Tamale Teaching Hospital were analyzed.

Table 1. Descriptive Statistics of CD4 count of Patients on Treatment

Variable	Frequency	Percent
Age(Years)		
14-29	80	26.6
30-49	197	65.7
50 +	23	7.7
Educational Level		
Basic	98	33
Nil	114	38
Sec/Tech	37	12
Tertiary	51	17
Number of Patient per Year		
2009	63	21
2010	85	29
2011	70	23
2012	73	24
2013	9	3
Gender		
Female	208	69
Male	92	31
Employment Status		
Employed	195	65
unemployed	105	35
Marital Status		
No	137	46
Yes	163	54
Religious Affiliation		
Christianity	103	34
Islam	197	66
Alcohol Consumption		
No	265	88
Yes	35	12
Smoking		
No	289	96
Yes	11	4
CD4 Count Status		
No	34	11
Yes	266	89
Regimen		
AZT300/3TC150/EFV600[a]	52	17
AZT300/3TC150/NVP200[b]	201	67
TDF300/3TC150/EFV600[c]	29	10
TDF300/3TC150/NVP200[d]	18	6

Note: a: Zidovudine/Lamivudine/ Efavirenz drug combination
b: Zidovudine/Lamivudine/Nevirapine drug combination
c: Tenofovir/Lamivudine/ Efavirenz drug combination
d: Tenofovir/Lamivudine/Nevirapine drug combination

The ages of the patients range from 14 - 50+ years with majority (66%) of patients in the 30 - 49 years bracket whereas the least (8%) was in the 50+ year group. The data revealed that the 30-49 years group is the most vulnerable and efforts at combating HIV and AIDS must be directed at this group. It also showed that introducing ART to this group will be most effective in realizing the purpose of the therapy. There were fewer males (31%) than females (69%) indicating again that female are the most vulnerable and must be targeted for effective therapy (ART). The mean change in CD4 cell count was 392. Eighty- nine percent (89%) of the patients had an increase of CD4 cell counts whiles eleven percent (11%) had no increment in CD4 cell counts. Thirty - eight percent (38%) of the patients had no formal education. Of the 62% who had formal education, 33% had basic education, 12% attained senior high school education (Technical school), and 17% had tertiary education. Large percentages (66%) of the patients were Muslims with 34% professing to be Christians. Fifty-four percent (54%) of the patients were married and forty - six percent (46%) were non- married. Also, 195 (65%) had some form of employment, whiles the rest were unemployed. For the type of treatment, sixty–seven percent (67%) were on Zidovudine/Lamivudine/Nevirapine, seventeen percent (17%) were on Zidovudine/Lamivudine/Efavirenz, ten percent (10%) and six percent (6%) were on Tenofovir/Lamivudine/Efavirenz and Tenofovir/Lamivudine/ Nevirapine respectively. Only few (12%) of the patients consumed alcohol perhaps because Tamale is predominantly a Muslim community. Furthermore only four percent (4%) of the patients smoked with majority (96%) not smoking.

3.2. Model Interpretation

It is expedient that practical inferences are drawn from fitted model and estimated regression coefficients to compare the difference levels of impact and effect among the variables. Holding other variables in the model constant for a unit change in the predictor variable, the difference in log-odd for positive outcome is expected to change through the respective coefficients. Predictors with positive coefficients will cause increase tendency for CD4 cell counts. Negative coefficients show decrease tendency for significant predictors. In fact, effective interpretation of the coefficients in logistic regression model depends on the ability to place meaning on the difference between two logits. The exponent of the difference (between two logits) gives the Odds Ratio defined as the ratio of the odds for the independent variable present to that for not being present.

3.3. Variable Selection

The significance of the association for each of the explanatory variables with the dependent variable was tested. Out of the eight (8) explanatory variables, two (age and gender) were statistically significant (had impact on CD4 cell counts) as shown in Table 2.

Table 2 provides the regression coefficient (β) Statistics test (to test the statistical significance) and the all-important Odds Ratio (Exp (β)) for each variable category.

Table 2. Parameter Estimation for the various variables

Variable	B[a]	S.E[b].	Wald[c]	Df[d]	Sig.[e]	Exp(B)[f]	95% C.I.for EXP(B)[g]	
							Lower	upper
age	0.055	0.024	5.241	1	0.022	1.057	1.008	1.108
gender(1)	1.020	0.421	5.852	1	0.016	2.772	1.213	6.332
employment(1)	0.113	0.417	0.074	1	0.786	1.120	0.494	2.538
education			5.124	3	0.163			
education(1)	1.170	0.596	3.854	1	0.050	3.221	1.002	10.355
education(2)	0.167	0.493	0.115	1	0.735	1.182	0.450	3.103
education(3)	0.764	0.699	1.192	1	0.275	2.146	0.545	8.449
marital(1)	0.524	0.411	1.619	1	0.203	1.688	0.754	3.780
religion(1)	-0.002	0.433	0.000	1	0.996	0.998	0.427	2.331
smoking(1)	-0.194	1.154	0.028	1	0.866	0.824	0.086	7.906
alcohol(1)	-0.006	0.692	0.000	1	0.993	0.994	0.256	3.860
regimen(1)	18.479	40192.746	0.000	1	1.000	1.060	0.000	0.000
regimen1	-19.609	40192.746	0.000	1	1.000	0.000	0.000	0.000
regimen2	-19.135	40192.746	0.000	1	1.000	0.000	0.000	0.000
regimen3	-0.989	1.198	0.681	1	0.409	0.372	0.036	3.893
Constant	-0.288	2.163	0.018	1	0.894	0.750		

Notes:
a: coefficient of the constant (also called the "intercept (β)") in the null model,
b: standard error around the coefficient for the constant,
c: Wald chi-square test that tests the null hypothesis that the constant equals 0,
d: degree of freedom for the Wald chi-square test
e: Score test is used to predict whether or not an independent variable would be significant in the model.
f: exponentiation of the B coefficient (an odds ratio). The value is given by default because odds ratios are easier to interpret than the coefficient which is in log-odds units
g: confidence interval: the range of values that we are confident that each of the odds ratio will lies.

The regression coefficient (β) the Wald statistic for the test statistical significance and the Odds ratio (Exp. β) for all the variables are shown in Table 2. Age and Gender had significant effect on the CD4 cell counts whiles the other variables were not significant in predicting CD4 cell counts. In terms of Gender, the Wald's test (*Wald=5.851, d f=1, p<0.016*) indicate significant effect. Similarly, the values for Age (*Wald=5.241, d f=1, p<0.022*) showed significant effect. Hence the logit model may thus be formulated from Table 2 g(x) = -0.288+0.055age+1.020gender.

Meanwhile Pseudo $R^2$of the Logit Regression Model consists of Cox & Snell R^2 and Nagelkerke R^2 because the

logistic regression does not have an equivalent of the R^2 in ordinary least square regression [25].

Table 3. Pseudo R Square of the Logit Regression Model for the CD4 cell Count

Step	-2 Log[a]	Cox & Snell R^2	Nagelkerke R^2
1	193.971	0.059	0.115

Notes: a: -2log likelihood.

193.971 compared to the -2LL for the null model in the 'omnibus test of model coefficients' prove that there was significant decline in the -2LL. The Cox & Snell R^2 value (0.059) and the Nagelkerke's R^2 (0.115) explain approximately the variations in the outcome. Hence, the R^2 values revealed 5.9% and 11.5% variations in the outcome by the Cox & Snell and Nagelkerke values respectively.

Furthermore, the Hosmer and Lemeshow test provides determination of the accuracy of the model fitting (Table). The null hypothesis is that the model provides good fit of the data and rejection of the null hypothesis is arrived only if the probability value is less than 0.05 (P<0.05) which is the confidence level.

Table 4. Hosmer and Lemeshow Test of the Logit Model for the CD4 cell counts

step	X^{2a}	b	P-value[c]
1	10.058	8	0.261

Notes: [b] degree of freedom; [c] probability value; [a] chi-square.

Table 4 shows a chi-square value of 10.058 with corresponding probability value of 0.261.The p-value (0.261) is greater than the level of significance (0.05) indicating the accuracy of fitting for the model. Omnibus Test of Coefficient is usually used to check that the new model is always an improved version over the baseline model [25]. The chi-square (X^2) is used to determine the difference between the Log-likelihood (-2LL) of the baseline model. With significantly reduced -2LL compared to the baseline model, there exist significant variance in the outcome of the new model indicating and is an improvement [25].

4. Discussion

The aim of the study was to investigate the determinants of the change in CD4 cell counts among patients on antiretroviral treatment (ART) at the Tamale Teaching hospital (TTH) of the Northern Region of Ghana. Reviews of medical records (300 files) of ART registered clients were analyzed. The results revealed that age and gender affected CD4 cell count response to ART. Furthermore, marital status, alcohol consumption, smoking, and employment status did not have effect on CD4 cell count response. The correlation between CD4 cell count response and gender was highly significant, indicated by the Wald statistic (5.852). The effect of Age on CD4 cell count response showed a factor of 0.016 (1.6%) [20]. Moreover, there was no evidence that alcohol consumption affected changes in CD4 cell count consistent with [22]. However, [23] showed that frequent alcohol use almost tripled the risk of drop in CD4 cell count response.

It is hoped that this work will convey essential message to policy makers to incorporate CD4 cell trends analysis studies (to aid CD4 cell count monitoring in ART centers). Since majority of the clients had substantial amount of CD4 count increment, it is imperative that clinicians should not delay commencing ART until the CD4 count falls far below 350cells/mm^3.

Subsequently, age affected CD4 cell counts response to ART; hence, detection of HIV infections at young age is paramount for early treatment when there the likelihood of significant CD4 cell counts increase. This will translate into increased immunity and prolong the lives of HIV patients.

5. Conclusion

The patients had their CD4 cell counts increased at certain points after initiation of treatment. The overall mean CD4 cell count was 391.67cell/mm^3. Only two variables (Age and Gender) were found to be significant and were included in the model ($g(x) = -0.288 + 0.55age + 1.020gender$). Furthermore, the level of significance of the regression coefficients for the two variables was less than 5% suggesting that these variables were indeed good predictors of CD4 cell counts. The results also showed that the model provided a reasonable statistical fit (chi-square value of 10.058 with corresponding probability value of 0.261 which is greater than the level of significance (0.05)). Since age affected (increase) in the CD4 cell counts in response to ART, detection of HIV infections at young age is paramount for early treatment when there the likelihood of significant CD4 cell count increase. This will translate into increased immunity and prolong the lives of HIV patients.

Ethical Considerations

Ethical approval was obtained from ethics committee of Tamale Teaching Hospital.

Statement of Competing Interest

The authors have no competing interests.

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