

Some Haematological Parameters in Human Immunodeficiency Virus (HIV) infected Africans: the Nigerian Perspective

O. Erhabor AMLS, AIBMS, BMLS, MSc, O. A. Ejele MBBS, MSc, DCP, FMCPATH, FWACP,

C. A. Nwauche BMED SC, MBBS, MSc, FWACP, F. I. Buseri AMLS, BMLS, MSc

Department of Haematology and Blood Transfusion, University of Port Harcourt Teaching Hospital, Port Harcourt, Nigeria.

ABSTRACT

Background: Haematologic abnormalities are among the most common manifestations of advanced human immunodeficiency virus (HIV) infection and acquired immunodeficiency syndrome (AIDS). A specific diagnosis of cause, severity and mechanism of cytopenia should be sought because of specific treatments or intervention may be indicated for its correction. This study was to determine some haematological parameters in HIV/AIDS infected Nigerians.

Method: One hundred HIV/AIDS infected previously antiretroviral naïve adult Nigerians, aged 18-58 year (males 47 and females 53) consisting of 88 symptomatic and 12 asymptomatic patients recruited into the antiretroviral pilot project in the Haematology department of the University of Port Harcourt Teaching Hospital between June 2002 to July 2003 were studied. Haematological parameters of hemoglobin, white cell count, platelet count, erythrocyte sedimentation rate and differential leucocyte count were determined. Data was analyzed using a multipurpose statistical package version 9 SPSS.

Result: The mean haemoglobin was 10.25 ± 1.97 g/dl (range 6.31-14.2 g/dl), severe anaemia occurred in 80% of subjects while 20% were non-anaemic. Haemoglobin values was found positively correlated to lymphocyte count ($r = .319$, $P = 0.01$) and inversely correlated to ESR ($r = -.343$, $P = 0.01$) and neutrophil count ($r = -.343$, $P = 0.01$). Red cell morphology was variable with majority normochromic and normocytic (64%) and 36% showing hypochromia and anisopoikilocytosis. The mean total WBC count was $4.51 \pm 1.82 \times 10^9/l$ (range 0.9-8.2 $\times 10^9/l$). Leucopaenia occurred in 10/100 (10%) of study population. Total white cell count showed a significant inverse correlation to lymphocyte count ($r = -.326$, $P = 0.01$). The mean neutrophil count was $2.32 \pm 1.58 \times 10^9/l$ (range 0.00-5.48).

Neutropaenia occurred in 24% of subjects. Neutrophil count showed a significant positive correlation with total white cell count ($r = .314$, $P = 0.01$) and a negative correlation with lymphocyte count ($r = -.982$, $P = 0.01$). Striking eosinophilia occurred in 3% of subjects. The mean platelet count was $170.07 \pm 49.03 \times 10^9/l$ (range 72-158 $\times 10^9/l$). Thrombocytopenia occurred in 10/100 (10%) of subjects. The mean erythrocyte sedimentation rate was higher than that in healthy Africans (mean 78.87 ± 39.33 mm fall/hour (range 0.2-158mm fall/hour).

Conclusion: Observation from this study will serve as a guide to clinicians caring for HIV patients in taking rational decision on haematological complications of HIV infection. This constitutes further evidence of the need for routine monitoring of some haematological parameters of HIV/AIDS infected Africans and before commencement of highly active antiretroviral therapy to ensure that mortality and morbidity are minimized and quality of life optimized.

KEY WORDS: Hematological Parameters; HIV; Nigerians; Africans.

Paper accepted for publication 24th November 2004.

INTRODUCTION

Globally the epidemic of HIV/AIDS is worse than ever with an estimated 33 million people living with the virus. Whereas the prevalence of HIV infection has increased only slightly in Europe and North America, there is an estimated 23 million HIV cases in Sub-Saharan Africa¹. The prevalence of HIV/AIDS has been increasing steadily in Nigeria from 1.8% in 1991 to 3.8% in 1993, 4.5% in 1996, 5.4% in 1999 and 5.8% in 2001².

Haematological abnormalities are among the most common manifestations of

HIV infection and AIDS. This is dominated by peripheral blood cytopenias. Anaemia has become more common with the advent of antiretroviral therapy and treatment for HIV related infections and malignancies³. Anaemia is a common manifestation of HIV disease⁴ and the anaemia is consistent with anaemia of chronic disease with a characteristics of anaemia with a mean haemoglobin of between 9 to 10g/dl⁵. According to the World Health Organization, haemoglobin value of 13.0 g/dl is the cut-off point below which anaemia is present at sea level for adult males and 12 g/dl for adult non-pregnant females⁶. Eight percent of HIV sero-positive patients, 20% with symptomatic middle stage disease and 71% of those with the Center for Disease Control (CDC) defined AIDS are anaemic⁷. Greater than 80% of symptomatic HIV/AIDS cases have a haematocrit of less than 30%, many of the patients have multiple peripheral blood cytopenia; 53% neutropaenia and 33% thrombocytopenia⁸.

Thrombocytopenia was first recognized as a manifestation of HIV/AIDS in homosexuals⁹ and drug abusers¹⁰. Thrombocytopenia defined as platelet count of $<150 \times 10^9/l$ has been found in 21% of patients with symptomatic AIDS and in 9% of asymptomatic HIV-sero-positive patients. Thrombocytopenia was found positively correlated with CD₄ count and older age¹¹. It has been established that with comparison with Caucasians, Africans have lower limit of platelet range. A lower limit of platelet range of $99 \times 10^9/l^{12}$ and $100 \times 10^9/l^{13}$ has been observed among healthy Nigerians. Platelet sequestration and destruction are the predominant mechanism of thrombocytopenia in early HIV infection and that decreased platelet production is the predominant mechanism of thrombocytopenia in advanced AIDS¹⁴.

It has been established that by comparison with Caucasians, healthy Africans have low total white cell count due to absolute neutropenia ascribed to genetics with nutrition as a co-factor. A mean total neutrophil count of $2.34 \times 10^9/l$ has been observed among healthy Nigerians¹⁵. It was previously thought that neutropenia was well tolerated in patients with AIDS but report suggests that the incidence of bacteria infection is higher in neutropaenic AIDS

patients compared to non-neutropaenic controls¹⁶. Although the haematological profile of HIV/AIDS infected individuals has been described in many centres in the West^{5,22}, there is paucity of data as regards African literature. This current research effort is focused on the identification, of haematological complications of HIV, in order to alert physicians caring for HIV infected Nigerians on the need to update information to make rational decisions on the haematological complications of HIV/AIDS and with the recent strategic decision by the Federal government to introduce her citizens living with HIV/AIDS to antiretrovirals, there is need to evaluate the haematological complications of HIV/AIDS patients at baseline to assess how highly active antiretroviral therapy (HAART) may further affect or improve cytopenias associated with HIV/AIDS to ensure that mortality and morbidity are minimized and quality of life and medical cost are optimized.

MATERIALS AND METHODS

One hundred previously antiretroviral naïve HIV sero-positive individuals recruited into the antiretroviral pilot study at the Haematology Department of the University of Port Harcourt Teaching Hospital (47 males and 58 females) aged between 19-58 years with 88 symptomatic and 12 asymptomatic constituted subjects for this study.

Specimen Acquisition and Laboratory Methods

Whole venous blood samples were collected by means of a 10ml hypodermic syringe into potassium EDTA anticoagulated tubes (5mls) and plain tubes without anticoagulant (5mls). Sera derived from the dry tubes were screened and confirmed for HIV using a double ELISA method made up of the World Health Organization (WHO) approved Immunocomb HIV 1& 2 kit (Organics, Israel) and Genscreen HIV 1 and 2 test kit (Bio Rad, France) in the absence of a western blot confirmatory test. Haematological parameters of haemoglobin was determined by the cyanmethaemoglobin method, total white cell count by Turks method, platelet count by the Breecher and Cronkite method of manual platelet count using ammonium oxalate, erythrocyte sedimentation rate by the Westergreen

method, blood films were made by the push-wedge technique, differential count done by the battlement method. Manual methods as described by Dacie and Lewis¹⁷ were used for all haematological investigations. Demographic data of age, sex and informed consent were obtained from all test subjects.

Statistical Analysis

Data was analyzed using a statistical package for personal computer (Version 9; SPSS Inc Chicago IL.). Statistical analysis of mean, standard deviation, Chi square test were used for discrete variables while correlations were compared by linear regression analysis. Differences were regarded as significant when $P \leq 0.05$.

RESULTS

The highest prevalence of HIV among subjects occurred in the 29-38 years age group $\frac{44}{100}$ (44%) while the lowest prevalence occurred in the 49-58 years age group $\frac{11}{100}$ (11%). Ninety-six percent of study subjects had HIV-1 infection, 2% had HIV-2 while 2% had dual HIV 1& 2 infection. Study subjects constituted of 53 females and 47 males representing a 1.1 male to female ratio. Table I show the HIV status based on age groups of study subjects.

The mean, standard deviations, ranges of the various haematological parameters and the level of haematological derangement observed among study subjects are shown in Tables II and III. The mean haemoglobin and ranges of study population was 10.25 ± 1.97 g/dl and 6.31 to

14.2g/dl. Anaemia, defined as haemoglobin value of <13g/dl for male and <12g/dl for females, occurred in 81% of subjects while 19% were non-anaemic. Blood film showed a varying degree in red cell morphology, with majority of subjects (64%) showing a normochromic and normocytic picture and 36% showing moderate anisopoikilocytosis and hypochromia. The mean total white blood cell count and range was $4.5 \pm 1.82 \times 10^9/l$ and $0.9-8.2 \times 10^9/l$ respectively. Leucopaenia defined as total WBC count was < $3.0 \times 10^9/l$ occurred in 10% of study subjects. Neutropaenia, defined as absolute neutrophil count of < $2.34 \times 10^9/l$ occurred in 24% of subjects. The mean neutrophil count was $2.32 \pm 1.58 \times 10^9/l$ and range of 0.00 to $5.48 \times 10^9/l$. Three (3) subjects had eosinophila with counts ranging from - 4.0 to $5.8 \times 10^9/l$. Thrombocytopaenia, defined as platelet count of < $100 \times 10^9/l$ occurred in 10% of study population. The mean ESR and range were 78.87 ± 39.33 mm/hr and 0.2 to 158mm/hr respectively. Haematological correlates in HIV infected study subjects are given in table IV. Haemoglobin value was found to be positively correlated with lymphocyte count ($r = .319$, $P < 0.01$) and inversely correlated with erythrocyte sedimentation rate (ESR) and neutrophil counts ($r = -.319$, $P < 0.01$) and ($r = .343$, $P < 0.01$) respectively. Total white cell count was found positively correlated with neutrophil count ($r = .314$, $P < 0.01$) and inversely correlated with lymphocyte count ($r = -.982$, $P < 0.01$).

Table I. Distribution of HIV Positivity Among Various Age Groups

Age Group (years)	No. & % HIV Positive	No & % HIV-1 Positive	No & % HIV-2 Positive	No & % HIV-1 & 2 Positive
19-28	22 (22%)	21 (95.5)	1(4.6)	-
29-38	44 (44%)	43 (97.7)	1 (2.3)	-
39-48	23 (23%)	22 (95.7)	-	1 (4.3)
49-58	11 (11%)	10 (90.9)	-	1 (9.1)

Table II. Mean Haematological Parameters and ranges of HIV infected subjects

Haematological Parameter	Mean	SD	Range
Haemoglobin (g/dl)	10.25	± 1.97	6.31 - 14.2
WBC (x 10 ⁹ /l)	4.51	± 1.82	0.9 - 8.2
Platelet (x 10 ⁹ /l)	170.07	± 49.03	72.0 - 268.1
ESR (mm/hr)	78.87	± 39.33	0.2 - 158
Neutrophils (x 10 ⁹ /l)	2.32	± 1.58	0.00 - 5.48
Lymphocyte (x 10 ⁹ /l)	2.18	± 1.01	0.16 - 4.20
Monocyte (x 10 ⁹ /l)	0.13	0.10	-2.0 - 3.8
Eosinophil (x 10 ⁹ /l)	0.17	0.19	-4.0 - 5.8

Table III. The incidence of Haematological Derangement Among HIV infected subjects

Haematological Derangement	Number of Study Subjects	% of Study Subject
Anaemia (Hb < 13g/dl male and <12 g/dl female)	80	80%
Leucopaenia (WBC < 3.0 x 10 ⁹ /l)	10	10%
Thrombocytopaenia (platelet < 100 x 10 ⁹ /l)	10	10%
Neutropaenia (Neutrophil < 2.34 x 10 ⁹ /l)	24	24%

Table IV. Haematological Correlates in HIV infected Subjects

	HB	WBC	PLAT	ESR	NEUT	LYMPH	MONO	EOSIN
HB	1.0001	-.120	-.171	-.319**	-.343**	.319**	.111	.151
WBC	-.120	1.000	-.094	-.001	.314**	-.326**	-.035	-.005
PLATELET	.171	-.094	1.000	-.083	-.029	.055	-.03	-.108
ESR	-.319**	-.001	-.083	1.000	.114	-.101	.020	-.081
NEUTROPHILS	-.343**	.314**	-.029	.114	1.000	-.982**	-.116	-.013
LYMPHOCYTES	.319**	-.326**	.055	-.101	-.982**	1.000	.028	-.013
MONOCYTES	.111	-.035	-.003	.020	-.116	.028	1.000	.140
EOSINOPHILS	.151	-.005	-.108	-.081	-.137	-.013	.140	1.000

* Correlation is significant at the 0.01 level (2-tailed). **Correlation is significant at the 0.05 level (2-tailed).

DISCUSSION

In this study we observed the highest HIV infection burden in subjects in the 29-38 years age group (44%), a 1:1 male to female ratio and a higher HIV-1 prevalence (96%) compared to 2% for HIV-2 and 2% for dual HIV-1 and 2 infections. These findings are in agreement with observations of other workers in Nigeria¹⁸⁻²⁰. These observation may have been accounted for by the fact that youths (29-38 years) are more sexually active and are more prone to high risk behaviour; maintenance of multiple sex partners, intravenous drug use and other high risk behaviour that makes them vulnerable. This observation spells doom for

Nigeria since the economically viable aspect of her population is worst hit.

In this present study we observed a mean haemoglobin value of 10.54 ± 1.96g/dl and a range of 6.3-14.2 g/dl. This value is however lower than that observed among healthy Nigerians^{12,21}. The anaemia seen in HIV infected subjects in this present study has the characteristics of anaemia of chronic infection with mean haemoglobin of 10 g/dl. This observation is in agreement with previous study by Perkocha *et al*⁵. Many factors have been incriminated in the etiology of anaemia associated with HIV infection. These include the direct myelosuppressive effect of HIV infection, the

secondary effect of drug therapy and other pre-existing or co-existing medical problems that may be prevalent in the HIV-infected population²². Other compounding factors responsible for the high incidence of anaemia in this study may be malnutrition, malaria and other parasitic infections. The incidence of anemia seen in this study (80%) is significantly lower than that obtained in previous study by Cosby *et al* (85%)⁸. The lower value seen in this study may have been due to the fact that unlike in the work of Cosby *et al*⁸, which involved symptomatic AIDS patients, ours included 12% of asymptomatic patients yet to present with AIDS defining symptoms. Haemoglobin value was found positively correlated with lymphocyte count $r = .319$, $p < 0.01$ and inversely correlated to ESR and neutrophil count $r = -.319$, $p < 0.01$ and $r = -.343$, $p < 0.01$ respectively: an indication that haemoglobin values in HIV infected Nigerians is directly proportional to lymphocyte count and inversely proportional to neutrophil count and erythrocyte sedimentation rate.

The mean total white cell count observed in this study $4.51 \times 10^9/l$ is at variance with that observed in healthy population in this environment. The lower limit of the total white blood cell range ($2.0 \times 10^9/l$) observed in this study is lower than that found in healthy Nigerians¹⁵. This observation may be due to suppression of the bone marrow resulting in abnormal granulopoiesis. The effect of antigranulocyte antibodies has been described in 30-67% of HIV infected²³ and the effect of medications used to treat HIV related opportunistic infections. White cell count in this study was found positively correlated with neutrophil count $r = -.326$, $p < 0.01$: an indication that WBC count is directly proportional to lymphocyte count. The incidence of neutropaenia seen in this study was 24%. This value is lower than that observed in a previous report⁸. The high incidence of neutropaenia seen in this study may be due to myelo-suppressive effect of drug therapy and the effect of antigranulocyte antibodies. Eosinophilia was present in 3% of study subjects. The high incidence of eosinophilia may have been due to the presence of tropical parasitic infections.

Thrombocytopenia was observed in 10% of HIV infected subjects in this study. We obtained a mean platelet count of $170.81 \pm 53.8 \times 10^9/l$ and a 95% platelet range of $72 - 268 \times 10^9/l$. The lower and upper limits of our platelet range is lower than that observed in healthy Nigerians^{12,13}. The incidence of thrombocytopenia obtained in this study involving 88 symptomatic and 12 asymptomatic HIV patient is however lower than that obtained among symptomatic AIDS patients in previous study⁸. Platelet sequestration and destruction has been suggested as the predominant mechanism of thrombocytopenia in early HIV infection¹⁴.

In this study we observed a mean ESR of 78.87 ± 39.33 and a 95% confidence interval ESR range of $0.2 - 158$ mm fall/hour. These values are however higher than that observed among healthy Nigerians in a previous study²¹. The rise in ESR may indicate some degree of anaemia and of the presence of a sub-clinical infection.

In this study we have observed a high prevalence of anaemia (80%), leucopaenia (10%), thrombocytopenia (10%) and neutropaenia (24%). This study indicates the need for routine haematological assessment of HIV infected Africans, before the commencement of highly active antiretroviral therapy. Observation from this study constitutes further evidence of the need for physicians caring for HIV/AIDS patients to update information required to make rational decisions concerning haematological complications of HIV/AIDS to ensure that mortality and morbidity are minimized and quality of life and medical cost are optimized.

REFERENCES

1. Satcher D. The global HIV/AIDS epidemic. JAMA 1999; 281:1479.
2. Summary findings from the 2001 HIV/syphilis sero-prevalence survey in Nigeria. Information for policy makers 2001: 30-50.
3. Yarchoan R, Mitsuya H, Myers C E, et al. Clinical Pharmacology of 3'-azido-2',3'-dideoxythymidine and related dideoxynucleosides. N Engl J Med 1989; 321:726-738.
4. Lundergreen JD. Comparison of long-term prognosis of patients with AIDS treated and not treated with Zidovudine. JAMA 1994; 271: 1088-1092.
5. Perkocha LA, Rodgers GM. Haematologic aspect of human immunodeficiency virus infection.

- Laboratory and clinical considerations. *Am J Hematol* 1988; 29:94-105.
6. De-Maeyer E, Adiels – Tegman M. The prevalence of Anaemia in the world. *World Health Statistics Quarterly* 1985; 8:302-316.
 7. Zon LI, Groopman JE. Haematologic manifestations of the Human Immunodeficiency Virus (HIV). *Br J Haematol* 1986; 66:251-256.
 8. Cosby C, Holzemer W C, Henry S B, *et al.* Haematological complications and quality of life in hospitalized AIDS patient care. *STD J* 2000; 4(5): 269-279.
 9. Walsh C, Krigel R, Lennette E, *et al.* Thrombocytopenia in a homosexual patient. Prognosis, response to therapy and prevalence of antibody to the retrovirus associated with Acquired Immunodeficiency Syndrome. *Ann Intern Med* 1985; 103:542-545.
 10. Savona S, Nardi MA, Lennette E, *et al.* Thrombocytopenic purpura in Narcotic addicts. *Ann Intern Med* 1985; 102:737-741.
 11. Peltier JY, Lambin P, Doinel C. Frequency and prognostic importance of thrombocytopenia in symptom free HIV-infected individuals. A five years prospective study. *AIDS* 1991; 5:381-384.
 12. Ukaejiofor E O, Isaacs-Sodeye WA, Seyide EA, *et al.* Normal haematological values in Adult Nigerians. *Niger Med J* 1979; 9(1):117-119.
 13. Essien E M, Usanga EA, Ayeni O. The normal platelet count and platelet factor 3. Availability in some Nigerian population groups. *Scand J Haematol* 1973; 10: 378-383.
 14. Najean PM, Rain JD. The mechanism of thrombocytopenia in patients with HIV infection. *J Lab Clin Med* 1994; 123:415-420.
 15. Ezeilo GC. The aetiology of neutropenia in healthy Africans. *East Afr Med J* 1974; 57: 936-942.
 16. Faber BF, Lesser M, Kaplan MH. Clinical significance of neutropenia in patients with immunodeficiency virus infection. *Infect Contr Hosp Epidemiol* 1991; 12: 429-434.
 17. Dacie JV, Lewis SM. *Practical Haematology*, 9th Ed. London: Churchill Livingstone, 2001: 19-56.
 18. Erhabor O. The geometrically increasing prevalence of HIV infection and its attendant social implications in Port Harcourt Nigeria. *Nig Lab J* 2001; 4(3): 27-29.
 19. Akinsete I, Akamu AS, Okany CC. Trends in HIV-sero-positivity among Visa Applicants in Lagos, Nigeria: A five year survey – 1992-1996. *Niger Postgrad Med J* 1998; 5 (2): 69-72.
 20. Ekweozor CC, Olaleye OD, Tomori O, *et al.* HIV sentinel surveillance in Ibadan/ Ogbomosho Zone of Oyo State. A pilot study. *Niger Med J* 1993; 24 (1): 1-4.
 21. Araba AB. A survey of Haematological variables in 600 healthy Nigerians. *Niger Med J* 1979; 9(1): 49-53.
 22. Evans RH, Scadden DT. Haematological aspects of HIV infection. *Best Prac Research Clin Haematol* 2000; 13 (2):215-230.
 23. Kimura S, Matasuda J, Ikematus S, *et al.* Efficacy of recombinant human granulocyte colony stimulating factor on Neutropenia in patients with HIV. *AIDS* 1990; 4:1251-12.