International Journal of Biosciences (IJB)
ISSN: 2220-6655 (Print) 2222-5234 (Online)
Vol. 2, No. 7, p. 11-22, 2012
http://www.innspub.net

RESEARCH PAPER OPEN ACCESS

Iron metabolism and antiretroviral therapy (ART) in women with HIV in Abidjan (Côte d'Ivoire)

Soualio Kamagate[1], Mathieu N. Bléyéré[1*], Mama Kone[1], Leandre K. Kouakou[1], Sylvie Doumate[2], Augustin K. Amonkan[3], Duni Sawadogo[4], Etienne E. Ehile[1], Henri Chenal[2], Paul A. Yapo[1]

[1]*University of Abobo-Adjamé*

[2]*Integrated Research Bioclinical Centre in Abidjan (IRBCA)*

[3]*University of Cocody*

[4]*University of Cocody*

Received: 15 June 2012
Revised: 29 June 2012
Accepted: 30 June 2012

Key words: Iron metabolism, HIV/AIDS, ART, women of reproductive age, Abidjan.

Abstract

We have initiated the investigations in women of reproductive age with HIV to evaluate and to characterize their iron metabolism. Moreover, to demonstrate the importance of ART, a study was conducted to compare the iron stores and the components of iron status between women of reproductive age with ART and those naive of ART. To do this, 60 women with ART and 60 women naive of ART were recruited in a specialized centre for treatment of HIV (ICBRA) based on the criteria for inclusion and exclusion. The mean age of women was 35.9 ± 0.4 years with extremes of 18 and 45 years. Blood samples were obtained from each subject to search the different biological indicators of iron status assessment. The results of our investigations showed that all the biological parameters searched of iron status evaluation, are degraded in enrolled women. Otherwise, our study revealed that women naive of ART indicated a more altered iron status than those with ART (88.3 % vs 78.3 %). Abnormal iron status consisted of iron deficiency, iron deficiency anaemia, inflammatory anaemia and inflammatory anemia associated with iron deficiency. Among the different components of iron status, inflammatory anaemia indicated high prevalence rates both in women naive of ART and women with ART (70 % vs 65 %). It appears from this study that antiretroviral treatment greatly disturbs iron metabolism in women of reproductive age with HIV infection. In addition, the inflammatory anaemia is significantly associated with ART in women with HIV infection.

*Corresponding Author: Mathieu N. Bléyéré ✉ bleyere@yahoo.fr

Introduction

More than a public health problem, HIV/AIDS is now a development problem and safety concern in the world. The fight against this pandemic is probably one of the major challenges of this century to ensure the harmonious development of our nations and to guarantee the living quality (UNAIDS/WHO, 2009). In Côte d'Ivoire, HIV prevalence in the population was 4.7 % according to the EIS in 2005 (UNGASS Country Report Cote d'Ivoire, 2010). It rose to 3.7 % in 2008 (UNAIDS/WHO, 2009). However, Côte d'Ivoire remains one of the most affected countries in the sub region of West Africa. Studies (UNGASS Country Report Cote d'Ivoire, 2010) were accurately estimated to 440000 the number of people living with HIV in Côte d'Ivoire since 2008 with 250000 women aged over 15 years and which remain the most concerned (6.9 % against 2.4 %). Despite this high prevalence, only 51820 subjects living with HIV were on antiretroviral therapy (ART). Antiretroviral therapy reveals many side effects often dramatic (Beuzit *et al*., 1992; Asuncion *et al*., 1998; Nguemaim *et al*., 2010). Investigations have shown that haematological complications are common in HIV infection (Fleming 1997; Massawe, 2002, Diallo *et al*., 2003, Nacoulma, 2007). Thus, HIV infection and antiretroviral therapy appear to be significant causes of anaemia incidence in women of reproductive age. And yet in Cote d'Ivoire, very few scientific works were performed on iron metabolism and antiretroviral therapy (ART) in women of reproductive age living with HIV/AIDS. This study aims to assess the iron status in these subjects in order to elucidate clearly the influence of antiretroviral drugs on the different biological indicators of iron metabolism. Moreover, our study intends to characterize possible changes in biological parameters of iron status evaluation among women of reproductive age on antiretroviral therapy (ART) and those naive to this treatment. In addition, our works will estimate different prevalences of iron deficiency and type of anaemias in these selected women. Otherwise, our study will compare the prevalence of the iron status components between the two groups of women and will indicate which one is most exposed to a possible alteration of iron metabolism during antiretroviral therapy (ART).

Materials and methods

Site and study population

This study was conducted from October 21, 2009 to December 21, 2010 in the Integrated Centre for Bioclinical Research of Abidjan (ICBRA). The study population consists of 120 HIV positive women aged 18 to 45 years and based on different social and professional groups. Women of reproductive age living with HIV type 1 represented for 99.2 % against 0.8 % infected with both HIV types 1 and 2. The study population has included 60 HIV positive women receiving antiretroviral therapy (ARV) and 60 HIV infected women naive of ART. These HIV positive subjects were screened through two types of HIV serology tests (Test Sets and Test Engineering II (HIV-1/HIV-2) to Integrated Centre for Bioclinical Research of Abidjan. After information and consent, 60 women came for voluntary testing, diagnosed with HIV and 60 women receiving antiretroviral therapy at least a year and have not developed complications of hypertension, diabetes, rheumatoid arthritis were included. The types of antiretrovirals commonly used by the subjects and prescribed after clinical trials are zidovudine, lamivudine and nevirapine (Coffi *et al*., 2010). However, women recently transfused and those who reported gastrointestinal and gynecological pathologies were excluded. In the same vein, women with menstruating were not retained. The women selected had a mean of 35.94 ± 0.39 years. The body mass index (BMI), number of pregnancies, childbirth and the period between births or pregnancies had respective mean values of 24.4 ± 0.4 $kg.m^{-2}$, 2.03 ± 0.1; 0.7 ± 0.04, 15.9 ± 1.2 weeks (Table 1). In the same table, our study has selected a low proportion (1.7 %) of adolescents compared to those whose age is physiologically normal (98.3 %). The body mass index was abnormal (insufficient and overweight) in 41.5 %

(Table 1). For our investigations, women were more multigravidae (67.5 %), multiparous (50 %) and more subjects with less than 36 months between pregnancies (74.2 %). In addition, women were well educated and have included married, single and some living in concubinage (Table 1).

Table 1. characteristics of study population.

Anthropometric and sociodemographic parameters	Total population N = 120	HIV positive women with ART N= 60	HIV positive women naïve ART N = 60
Age (Years)			
18 – 19	35,9 ± 0,5	35,73 ± 0,8	36,15±0,8
20 – 45	2/(1,7 %)	1/(1,67 %)	1/(1,67 %)
	118/(98,3 %)	59/(98,3 %)	59/(98,3 %)
BMI (kg.m^{-2})			
< 19,8	24,4 ± 0,5	23,5 ± 0,6	25,4 ±0,8
19,8 – 26	18/(15 %)	9/(15 %)	9/(15 %)
> 26	66/(55 %)	39/(65 %)	27/(45 %)
	36/(30 %)	12/(20 %)	24 /(40 %)
Gestity			
Primigravidae	2,02 ± 0,2	1,8 ± 0,3	2,3 ± 0,3
Multigravidae	39/(32,5 %)	15/(25 %)	24/(40 %)
	81/(67,5 %)	24/(40 %)	57(/(95 %)
Parity			
Nulliparous	0,7 ± 0,04	0,65 ± 0,1	0,7 ± 0,1
Primiparous	39/(32,5 %)	21/(35 %)	18/(30 %)
Multiparous	21/(17,5 %)	15/(25 %)	6/(10 %)
	60/(50 %)	24/(40 %)	36/(60 %)
Space between births (Months)			
< 36	15,9 ± 1,2	13,2 ± 2,3	18,7±2,4
≥ 36	90/(75 %)	47/(78,3 %)	43/(71,7 %)
	30/(25 %)	13/(21,7 %)	17/(28,3 %)
Educational attainment			
Uneducated	27/(22,5 %)	10/(16,7 %)	17/(28,3 %)
primary school	24/(20 %)	12/(20 %)	12/(20 %)
Secondary school	40/(33,3 %)	26/(43,33 %)	14/(23,3 %)
Superior	31/(26,7 %)	15/(25 %)	17/(28,3 %)
Matrimonial status			
Married	35/(29,2 %)	19/(31,7 %)	16/(26,7 %)
Singles	48/(40 %)	28/(46,7 %)	20/(33,3 %)
Concubinage	37/(30,8 %)	13/(21,7 %)	24/(40 %)
widowers	-	-	-

N: Total number of each subject group; ART: antiretroviral theraty.

Blood samples and assays of biological parameters

At each of the women recruited, a blood sample collected in dry tubes and tubes containing an anticoagulant with 5 ml for each, was carried out at the elbow fasting in the morning. Whole blood collected in tubes with anticoagulant (EDTA) has achieved the measurement of CD4 lymphocytes (by flow cytometry with Fascalibur ®) and blood count by the automatic Sysmex XT 2000i. The blood in dry tubes was centrifuged at 3000 g/min for 3 min to obtain serum. The resulting serum was used to determine HIV status and biochemical indicators of iron status assessment. For HIV status, the most used in the care centres is using of two successive tests. Once the first test (Determine) is positive, we proceed to discrimination test (Genie II HIV-1/HIV-2) to determine the type of HIV. The quantitative determination of biochemical parameters (serum iron, serum transferrin and serum ferritin) in human serum is based on a colorimetric technique available on most automated COBAS

INTEGRA 400. For this determination, the COBAS INTEGRA kits Iron (IRON), Tina-as Transferrin ver.2 (TRSF2) TRSF2 Test, test and Ferritin Gen.2 ID 0-567 (FERR2) FERR2 Test, test ID 0-078, containing *in vitro* diagnostic reagents were used. Each dose of the blood sample from the same collection is duplicated to reduce the errors of manipulation. And the mean of the two obtained is used for the study. The total iron binding capacity (TIBC) and the saturation coefficient of transferrin (SCT) were obtained by calculations.

Table 2. Mean values of biological parameters.

Haematological parameters	Total population N = 120	HIV positive women with ART N= 60	HIV positive women naïve ART N = 60	p-values	Reference values[α]
Red blood cells count					
Red blood cells (10^{12}/l)	3.9 ± 0.1	3.8 ± 0.1	4 ± 0.1	0.9 (NS)	4-5.4
Hemoglobin (g /dl)	11.2 ± 0.2	11.7 ± 0.2	10.7 ± 0.2	0.7 (NS)	12-16
Hematocrit (%)	35.3 ± 0.4	36.4 ± 0.4	34.1 ± 0.6	**0.03 (S)**	40-54
Erythrocyte indices					
MCV (fl)	91.7 ± 1.1	96.2 ± 1.5	87.3 ± 1.3	0.3 (NS)	80-100
MCH (pg)	29.1 ± 0.4	30.9 ± 0.6	27.3 ± 0.5	0.05 (NS)	27-31
MCHC (g/dl)	31.7 ± 0.1	32.1 ± 0.2	31.3 ± 0.2	0.4 (NS)	32-36
Plasma compartment					
Serum iron (µmol/l)	11.8 ± 0.6	13.8 ± 0.7	9.5 ± 0.6	0.9 (NS)	6.6-26
Serum transferrin (g/l)	2.5 ± 0.1	2.6 ± 0.1	2.4 ± 0.1	0.9 (NS)	2-3.2
TIBC (µmol/l)	61.6 ± 1.4	64.4 ± 1.7	58.9 ± 2	0.8 (NS)	50-90
SCT (%)	10.9 ± 0.5	12.6 ± 0.7	9.2 ± 0.5	0.6 (NS)	15-35
Iron store compartment					
Serum ferritine sérique (µg/l)	161.2 ± 22	116.4 ± 23.1	206 ± 36.7	**3.10^{-7} (S)**	15-150

N: Total number of each subject group; n: subject number observed in each group; ART: antiretroviral therapy; MCV: Mean corpuscular volume; MCH: Mean corpuscular hemoglobin; TIBC: Total iron binding capacity; SCT: Saturation coefficient of transferrin; S: Statistically different for p value < 0.05; NS: Not statistically significant for p value > 0.05; α: Haemotological reference parameters respectively in boys and girls according to Vernet *et al.* (2001)

Assessment and statistical analysis of biological parameters

To better appreciate the parameters of our laboratory testing, conventional criteria were selected. They associated the recommendations of international organizations (WHO), the French Society of Clinical Biology (SFBC/France), French Society of Hematology (SFH/France-Group of Cellular Haematology), the Society of Nutrition and Diet of the French Language (France), Centre for Disease Control and Prevention (WHO/CDCP) and the Institute of Medicine (IOM/US) (Vernet *et al.*, 2001; IOM/US, 1990, UNICEF/UNU/WHO, 2001; SNDLF, 2001). The mean values of biological parameters obtained were submitted to a Student's t test for independent samples with the computer program Statistica Statsoft Windows version 7.1 (Statsoft, 2005) in order to evaluate the influence of antiretroviral therapy on iron metabolism. The different observed proportions for biological indicators of iron status were compared by the likelihood test or G test log likehood ratio with the software version R.2.0.1 Windows (Ihaka and Gentleman, 1996). The statistical significance was defined for a p-value less than 0.05.

RESULTS

Changes in biological parameters between different groups of women

The mean values of blood counts parameters were all abnormal except the red blood cells in women without antiretroviral treatment (Table 2). At the level of erythrocyte indices, the mean values were all normal

except the mean corpuscular hemoglobin concentration (MCHC) in all subjects and in women naïve of antiretroviral therapy (Table 2). For biochemical indicators, the mean values were normal except for the saturation coefficient of transferring (below the norm) and serum ferritin which is well above the norm for all subjects and in women of reproductive age naïve of antiretroviral treatment (Table 2).

Table 3. Compared proportions of erythrocyte parameters.

Erythrocyte parameters	Total population N = 120	HIV positive women ART N = 60	HIV positive women naïve ART N = 60	p-values
	n/% (IC 95 %)	n/% (IC 95 %)	n/% (IC 95 %)	
Hemoglobin (g/dl)				
< 12	81/67.5 (55.5-79.5)	34/56.7 (44.2-69.2)	47/78.3 (67.9-88.7)	0.05 (NS)
12-16	39/32.5 (26.8-38.2)	26/43.3 (30.8-55.8)	13/21.7 (11.3-32.1)	0.007 (S)
Hematocrit (%)				
< 40	109/90.8 (74.6-107)	55/91.7 (84.7-98.7)	54/90 (67.4-112.6)	1 (NS)
40-54	11/9.2 (7.6-10.8)	5/8.3 (1.3-15.3)	6/10 (7.6-12.4)	1 (NS)
MCV (fl)				
< 80	17/14.2 (8-20.4)	4/6.6 (0.3-12.9)	13/21.7 (11.3-32.1)	0.004 (S)
80-100	70/58.3 (49.5-67.1)	43/71.7 (60.3-83.1)	27/45 (32.4-57.6)	0.01 (S)
100 -114	33/27.5 (19.5-35.5)	13/21.7 (11.3-32.1)	20/33.3 (21.4-45.2)	0.05 (NS)
MCH (pg)				
< 27 ou > 31	84/70 (61.8-78.2)	30/50 (37.3-62.7)	54/90 (82.4-97.6)	0.0007 (S)
27-31	36/30 (21.8-38.2)	30/50 (37.3-62.7)	6/10 (2.4-17.6)	7.10^{-7} (S)
Types of anaemias				
NNA	27/22.5 (18.6-26.4)	10/16.7 (12.6-20.8)	17/28.3 (21.3-35.4)	0.5 (NS)
NHA	21/17.5 (14.5-20.5)	4/6.7 (5.1-8.2)	17/28.3 (21.3-35.4)	0.009 (S)
MHA	15/12.5 (10.4-14.6)	5/8.3 (10.3-6.4)	10/16.7 (12.6-20.8)	0.08 (NS)
mHA	18/15 (12.4-17.6)	8/13.3 (10.1-16.6)	10/16.7 (12.6-20.8)	0.88 (NS)

N: Total number of each subject group; n: subject number observed in each group; ART: antiretroviral therapy; MCV: Mean corpuscular volume; MCH: Mean corpuscular hemoglobin; %: Proportion indicated in each subjects group; S: Statistically different for p value < 0.05; NS: Not statistically significant for p value > 0.05; NNA: normocytic normochromic anaemia; MHA: microcytic hypochromic anaemia; NHA: normocytic hypochromic anaemia; mHA: macrocytic hypochromic anaemia.

According to Table 2, no significant differences were observed between the different groups of women concerning the mean values of red blood cells, hemoglobin, mean corpuscular volume (MCV), the mean corpuscular hemoglobin (MCH), the mean corpuscular hemoglobin concentration (MCHC), serum iron, serum transferrin, the total iron binding capacity and coefficient saturation of transferrin ($p > 0.05$). However, the hematocrit decreased in women of reproductive age naive of antiretroviral treatment compared to other women ($p = 0.03$). This table also reported a highly significant increase of serum ferritin in women of reproductive age naive of antiretroviral treatment compared to those receiving antiretroviral therapy ($p = 3.10^{-7}$).

Distribution of biological parameters proportions and components of iron status

Table 3 compares the different proportions of red cell parameters between HIV infected women receiving antiretroviral therapy and HIV infected women naive of antiretroviral treatment. The results showed high rates of anaemia (56.7 % vs 78.3 %), normal rates of mean corpuscular volume (71.7 % vs 45 %), of hypochromia (50 % vs 90 %) in both groups of women. Furthermore, no significant differences ($p > 0.05$) were observed between the two groups of subjects on the

rates of anaemia and hematocrit. The results of the study also revealed that the rates of microcytosis (6.6 % vs 21.7 %) and macrocytosis (21.7 % vs 33.3 %) were higher in women naïve of antiretroviral treatment than in women receiving antiretroviral therapy. However, many women receiving treatment showed normal values of mean corpuscular volume (71.1 % vs 45 %) compared to naive women (p = 0.01).

Table 4. Compared biochemical parameters proportions between the two groups of women.

Biochemical parameters	Total population N = 120	HIV-PW with ART N = 60	HIV-PW naïve ART; N = 60	p-values
	n/% (IC 95 %)	n/% (IC 95 %)	n/% (IC 95 %)	
Serum Iron (µmol/l)				
< 6.6	18/15 (12.4-17.6)	2/3.3 (2.6-4)	16/26.7 (20.1-33.3)	$5.1.10^{-6}$ (S)
6.6-26	99/82.5 (61.8-103.2)	56/93.3 (69.8-116.8)	43/71.7 (53.7-89.7)	0.1 (NS)
> 26	3/2.5 (2.2-2.8)	2/3.3 (2.6-4)	1/1.7 (1.4-2)	0.5 (NS)
Serum transferrin (g/l)				
< 2	19/15.8 (13.1-18.5)	5/8.3 (6.3-10.3)	14/23.3 (8.2-29.1)	0.006 (S)
2-3.6	96/80 (65.8-94.2)	52/86.7 (64.9-108.5)	44/73.3 (54.9-91.7)	0.3 (NS)
> 3.6	5/4.2 (3.5-4.9)	3/5 (3.9-6.1)	2/3.3 (2.6-4)	0.6 (NS)
TIBC (µmol/l)				
< 50	19/15.8 (13.1-18.5)	5/8.3 (6.3-10.3)	14/23.3 (8.2-29.1)	0.006 (S)
50-90	96/80 (65.8-94.2)	52/86.7 (64.9-108.5)	44/73.3 (54.9-91.7)	0.3 (NS)
> 90	5/4.2 (3.5-4.9)	3/5 (3.9-6.1)	2/3.3 (2.6-4)	0.6 (NS)
SCT (%)				
< 15	96/80 (65.8-94.2)	42/70 (52.4-87.6)	54/90 (82.4-97.6)	0.1 (NS)
15-35	24/20 (16.5-23.5)	18/30 (22.5-37.5)	6/10 (7.1-12.9)	0.001 (S)
Serum ferritin (µg/l)				
< 15	15/12.5 (10.2-14.8)	7/11.6 (8.8-14.4)	8/13.3 (4.7-21.9)	0.7 (NS)
15-150	73/60.8 (50-71.6)	40/66.7 (49.9-83.5)	33/55 (42.4-67.6)	0.3 (NS)
> 150	32/26.7 (22-31.4)	13/21.7 (13.3-27.1)	19/31.7 (19.9-43.5)	0.2 (NS)
NIS	20/16.7 (10-23.4)	13/21.7 (11.3-32.1)	7/11.7 (3.6-19.8)	0.1 (NS)
AIS	100/83.3 (76.6-90)	47/78.3 (67.9-88.7)	53/88.3 (80.2-96.4)	0.4 (NS)
ID	9/7.5 (2.8-12.2)	6/10 (2.4-17.6)	3/5 (0.5-10.5)	0.2 (NS)
IDA	6/5 (1.1-8.9)	0/0 (0-0)	6/10 (2.4-17.6)	0.0001 (S)
IA	81/67.5 (59.1-75.9)	39/65 (52.9-77.1)	42/70 (58.4-81.6)	0.7 (NS)
IA+ ID	4/3.3 (0.1-6.5)	2/3.3 (1.2-7.8)	2/3.3 (1.2-7.8)	1 (NS)

HIV-PW: HIV-positive women; N: Total number of each subject group; n: subject number observed in each group; ART: antiretroviral therapy; %: Proportion indicated in each subjects group; TIBC: Total iron binding capacity; SCT: Saturation coefficient of transferrin; S: Statistically different for p value < 0.05; NS: Not statistically significant for p value > 0.05; NIS: Normal iron status; AIS: Abnormal iron status; ID: Iron deficiency; IDA: Iron deficiency anaemia; IA: Inflammatory anaeamia.

The combination of several haematological parameters summarized in Table 3 (hemoglobin, mean corpuscular volume and mean corpuscular hemoglobin) indicated in selected women the prevalences of normocytic normochromic anaemia (NNA), normocytic hypochromic anaemia (NHA), microcytic hypochromic anaemia (MHA) and macrocytic hypochromic anaemia (mHA). Normocytic hypochromic anaemia was significantly (p = 0.009) higher in women naive to antiretroviral treatment (28.3 %) compared to women on ART (6.7 %).

Table 5. Iron status and progression of HIV infection.

Components of iron status and CD4 count		Total population N = 120	HIV positive women with ART N = 60	HIV positive women naïve ART N = 60	p-values
Taux de CD4 (cellules/mm³)		n (%)	n (%)	n (%)	
< 200		34 (28.3)	8 (13.3)	26 (43.3)	**$4.3.10^{-5}$ (S)**
200-499		59 (49.2)	34 (56.7)	25 (41.7)	0.1 (NS)
≥ 500		27 (22.5)	18 (30)	9 (15)	**0.02 (S)**
Iron status	**Stages of infection**				
Normal status	A	5 (4.2)	3 (5)	2 (3.3)	0.6 (NS)
	B	8 (6.7)	6 (10)	2 (3.3)	0.06 (NS)
	C	6 (5)	3 (5)	3 (5)	1 (NS)
Iron deficiency	A	3 (2.5)	3 (5)	0 (0)	**0.008 (S)**
	B	5 (4.2)	3 (5)	2 (3.3)	0.6 (NS)
	C	1 (0.9)	0 (0)	1 (1.7)	0.12 (NS)
Iron deficiency anaemia	A	-	-	-	-
	B	4 (3.4)	1 (1.7)	3 (5)	0.19 (NS)
	C	3 (2.5)	0 (0)	3 (5)	**0.008 (S)**
Anémie inflammatoire	A	17 (14.2)	12 (20)	5 (8.3)	**0.03 (S)**
	B	42 (35)	22 (36.7)	20 (33.3)	0.68 (NS)
	C	21 (17.5)	5 (8.3)	16 (26.7)	**0.001 (S)**
Inflammatory anaemia + Iron deficiency	A	1 (0.9)	1 (1.7)	0 (0)	0.12 (NS)
	B	1 (0.9)	1 (1.7)	0 (0)	0.12 (NS)
	C	3 (2.5)	0 (0)	3 (5)	**0.008 (S)**

N: Total number of each subject group; n: subject number observed in each group; ART: antiretroviral therapy; %: Proportion indicated in each subjects group; S: Statistically different for p value < 0.05; NS: Not statistically significant for p value > 0.05.

At the level of plasma compartment, highly significant differences ($p < 0.01$) were observed between the two women groups for proportions of serum iron, serum transferrin, total iron binding capacity for the values below the normal and saturation coefficients of transferrin with values between 15 % and 35 %. No other significant difference ($p > 0.05$) was shown between these two groups of women even in iron stores (serum ferritin). In all, women of childbearing age with HIV infected and naive of antiretroviral treatment presented the highest pathological proportions except at normal values of the saturation coefficient of transferrin (Table 4).

The results of our work has shown in the components of iron status, the presence of normal iron status, iron deficiency, iron deficiency anaemia, inflammatory anaemia and inflammatory anaemia with iron deficiency (Table 4). Moreover, no significant difference ($p > 0.05$) was observed between the two groups of women. However, women receiving antiretroviral therapy have not indicated iron deficiency anaemia (0 %) compared with women naive of this treatment (10 %, $p = 0.0001$). Moreover, the prevalence of inflammatory anaemia was very high in two groups of subjects. In addition, abnormal iron status was higher among women naive of antiretroviral therapy (88.3 %) against 78.3 % in women on ART (Table 4).

Prevalence of iron deficiency and types of anaemias according to HIV infection progression

The proportions distribution of women with HIV by the rate of CD4 listed in Table 5 showed an immunodeficiency highly significant ($p = 4.3.10^{-5}$) in subjects without antiretroviral therapy compared to women on ART. In the same table, the prevalence of CD4 above normal was significantly ($p = 0.02$) higher

in women of reproductive age on ART than in those naive to antiretroviral therapy. As for the normal values of CD4, no significant difference ($p > 0.05$) was observed between the two groups of women (Table 5). Analysis of the results obtained from the classification based on HIV infection progression and components of iron status showed that the inflammatory anaemia was severely present in both groups of women. In stage A, it was significantly ($p = 0.03$) more prevalent among women on ART than women naive to ART (20 % vs 8.3 %). At stage B, although being the high proportion among both women on ART (36.7 %) and women naive to ART (33.3 %) no significant difference was observed ($p > 0.05$). In contrast, in stage C, it was significantly ($p = 0.001$) prevalent in women naive to ART than in those on ART (Table 5). In addition, highly significant differences ($p < 0.01$) were indicated between the two groups of women for iron deficiency in stage A (5 % vs 0 %), for iron deficiency anaemia at stage C (0 % vs 5 %) and for inflammatory anaemia associated with iron deficiency at stage C (0 % vs 5 %).

Discussion

The study that we initiated, include aspects physiology of nutrition, HIV infection and antiretroviral therapy (zidovudine, lamivudine and nevirapine). The aspect physiology of nutrition concerns iron metabolism. The metabolic pathways of this micronutrient affect all major physiological functions of the body (Brock *et al.*, 1994). The disruption of its metabolism leads to dysfunction in populations even in good health (Gordeuk *et al.*, 2001). Iron deficit and overload are the real problems in subjects infected in general and especially those infected with HIV. They use antiretroviral therapy to improve their defense system by increasing the rate of CD4 (Beuzit *et al.*, 1992). This result is reported in our investigations among women of childbearing age receiving antiretroviral therapy compared with those naive to this treatment. Antiretroviral drugs constitute a set of anti-infective medicines active on the viruses acquired immunodeficiency syndrome (HIV-1 and HIV-2). These are drugs mainly acting virostatics usually through enzyme inhibition. Antiretroviral therapy is palliative solution currently used (Potdar *et al.*, 2011). It reduces the viral load and increases the CD4 cell counts, allowing the survival and improvement quality life of those infected with HIV.

However, it involves more trouble within the body that receives (Danwe *et al.*, 2005; Ngondi *et al.*, 2007; Mobarak, 2011). This is the case of anaemia that is common in subjects receiving antiretroviral therapy (Drain *et al.*, 2007). The results of our study support this finding in our subjects receiving antiretroviral therapy that indicate a high rate of anaemia (hemoglobin < 12 g/dl) of 56.7 %. Conversely, our observation is contrary to the works of Moh *et al.* (2005) and Johannessen *et al.* (2011) in Côte d'Ivoire and among Tanzanian HIV infected who presented respectively lower prevalences of anaemia from 31 % to 17 % and 77.4 % to 39.1 %. Anaemia is caused by several factors. All the subjects of our study are already infected with HIV and some are on antiretroviral treatment, the main reason in our study may be food. In this sense, an evaluation and a characterization of the different components of iron status in women with HIV receiving and not antiretroviral therapy were performed by determining of biological indicators of iron metabolism. The major observation of this study is the very high prevalence of abnormal iron status as indicated both in the two groups of selected women (78.3 % vs 88.3 %). Women without ART showed an altered iron status more than those on treatment. Several studies have indicated that in populations living with HIV/AIDS, iron metabolism is significantly degraded (Trumbo *et al.*, 2001; Omoregie *et al.*, 2009, Russell *et al.*, 2010). This mechanism involves a recently identified protein, hepcidin which prevents the export of iron out of cells of duodenal and reticuloendothelial system, but not measured in clinical practice (Handelman and Levin, 2008). Furthermore, the synthesis of ferritin is directly increased by inflammation, independently and beyond

the level of iron stores. Ferritin does not therefore reflect more strictly the reserve of iron in the body in this situation (Boelaert *et al*., 1996, Semba *et al*, 2002). In addition, antiretroviral therapy increases CD4 cell counts as seen in the group of women on zidovudine, lamivudine and névirépine. However, during the progression of HIV infection, iron deficiency, iron deficiency anaemia and inflammatory anaemia are reported. These same results are shown in Gambia and Mexico (McDermid *et al*., 2007; Mata-Marín, 2010).

Conclusion

At the end of our study, women of reproductive age in Abidjan with HIV showed a strongly altered iron metabolism. All biological indicators assessment of iron status have changed significantly. Iron deficiency, iron deficiency anaemia and inflammatory anaemia are observed in both groups of selected women. This study also reveals that women naive of antiretroviral presented the higher rate of abnormal iron status than women on antiretroviral treatment. Among the different components of iron status, inflammatory anaemia is the most observed in all subjects. However, women naive to antiretroviral therapy have the proportions of pathological iron metabolism most pronounced. Antiretroviral therapy increases CD4 cell counts in treated contrary to the naive. HIV and its therapy are causing complications during infection as the persistence of anaemia (inflammatory anaemia). Antiretroviral therapy should be monitored in subjects infected with HIV with the achievement of regular laboratory tests for controlling the associated complications with this type of treatment. Among these analyzes, evaluation of the metabolism of nutrients (macronutrients and micronutrients) and nutritional assessment are necessary in populations infected with HIV. In the same direction, the determination of hepcidin and all the mechanisms of inflammation in people living with HIV need to be elucidated. Moreover, supplementation with micronutrients such as iron, zinc, copper, folic acid and serial vitamins B is essential in order to avoid the early onset of anaemia in subjects infected with HIV and also on antiretroviral therapy.

Acknowledgements

We wish to thank the managers and staff from the Integrated Centre for Bioclinical Research of Abidjan (ICBRA) for accepting such a study in their structure in cooperation with the University of Abobo-Adjame. Our appreciation is also due to Dr. Leonie Kouonon Clemence and Mr Barnabé Gouzilé respectively from the University of Abobo-Adjame and the Regional Unit for Higher Education in Korhogo for their guidance during the drafting of this publication.

Abbreviations

ART: Antiretroviral therapy; TIBC: Total Iron Binding Capacity; SCT: Saturation Coefficient of Transferrin; NIS: Normal Iron Status; ID: Iron Deficiency; IDA: Iron Deficiency Anaemia; IA: Inflammatory Anaemia; IA+ID: Inflammatory Anaemia associated with Iron Deficiency; S: Statistically different for p value < 0.05; NS: Not statistically significant for p value > 0.05; N: Total number of each subject group; SEM: Standard error of mean;

Funding

This research did not receive any specific grant from any funding agency in the public, commercial, or not-for-profit sector.

Authors' contributions

All authors contributed equally in the study. They made substantial contributions to the design of the study, the collection of the data as well as the preparation and analysis of the data. They also drafted the manuscript and gave final approval for its submission to the journal for consideration of publication.

Declaration of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

References

Asuncion G, Del Olmo ML, Sastre J, Millan A, Pellin A, Pallardo FV. 1998. AZT treatment induces molecular and ultrastructural oxidative damage to muscle mitochondria. Prevention by antioxidant vitamins. Journal of Clinical Investigation **102**, 4–9.

Beisel WR. 1992. History of nutritional immunology: introduction and overview. Journal Nutrition **122**, 591–6.

Beuzit Y, Bougarel J, Ngouonimba J. 1992. Modifications hématologiques périphériques et médullaires lors de l'infection par le Virus de l'Immunodéficience Humaine (VIH) en Afrique centrale. Medecine Tropicale **40**, 193-199.

Boelaert JR, Weinberg GA, Weinberg ED. 1996: Altered iron metabolism in HIV infection: mechanisms, possible consequences, and proposals for management. Infectious agents and disease **5(1)**, 36-46.

Brock JH. 1994. Iron in infection, immunity, inflammation and neoplasia. In: Iron Metabolism in Health and Disease. Ed. Brock JH, Halliday JW, Pippard MJ, Powell LW.Philadelphia: Saunders, 353–389.

Bruera D, Luna N, David DO, Bergoglio LM, Zamudio J. 2003. Decreased bone mineral density in HIV-infected patients is independent of antiretroviral therapy. AIDS **17**, 1917–1923.

Chevalier P, Sevilla R, Sejas E, Zalles L, Belmonte G, Parent G. 1998. Immune recovery of malnourished children takes longer than nutritional recovery: implications for treatment and discharge. Journal of Tropical Pediatrics **44(5)**, 304–7.

Coffie PA, Tonwe-Gold B, Tanon AK, Amani-Bosse C, Bédikou G, Abrams EJ, Dabis F, Ekouevi DK. 2010. Incidence and risk factors of severe adverse events with nevirapine-based antiretroviral therapy in HIV-infected women. MTCT-Plus program, Abidjan, Cote d'Ivoire BMC Infectious Diseases **10**, 188.

Danwe C, Atchou G, Nkam M, Mbuagbaw J, Mougnoutou R, Nkouanfack C, Mbanya D, Agbor AG, Lohoue J, Ngogang YJ. 2005. Effect of Antiretroviral Therapy on Lipid Metabolism in HIV/AIDS Subjects in Cameroon. Journal of Medical Sciences **5**, 78-82.

Diallo D A, Baby M, Dembele M, Keita A, Sidibe A, Cisse IAH, Diop CT, Maïga II, Traore AK, Traore HA. 2003. Fréquence, facteurs de risque et valeur pronostique de l'anémie associée au au VIH/sida chez l'adulte au Mali. Bulletin de la Société de Pathologie Exotique **96 (2)**, 123-127.

Drain PK, Kupka R, Mugusi F, Fawzi WW. 2007. Micronutrients in HIV-positive persons receiving highly active antiretroviral therapy. American Journaj of Clinical Nutrition **85**, 333– 45.

Fleming A F. 1997. HIV and blood transfusion in sub-Saharan Africa. Transfusion science **18**, 167-79.

Gordeuk VR, Delanghe JR, Langlois MR, Boelaert JR. 2001. Iron status and the outcome of HIV infection: an overview. Journal Clinical Virology **20(3)**, 111-5.

Handelman, GJ, Levin NW. 2008. Iron and anemia in human biology: a review of mechanisms. Heart Failure Reviews **13(4)**, 393-404.

Ihaka R, Gentleman R. 1996. R : a language for data analysis and graphics. J. computation, graphic and statistic **5**, 299-314.

Institute of Medecine (IOM/USA). 1990. Comittte on nutritional status during pregnancy and lactation. Nutrition during pregnancy: weight gain and nutrient supplements. Washington DC. National Academy Press.

Johannessen, A., E. Naman, S.G. Gundersen and J.N. Bruun, 2011. Antiretroviral treatment reverses HIV-associated anemia in rural Tanzania. BMC Infect Dis.

Katodritou, E. and J. Christakis, 2006. Recent advances in the pathogenesis and management of anaemia of chronic disease. Haema **9(1)**, 45-55.

Kupka R, Msamanga GI, Mugusi F, Petraro P, Hunter DJ, Fawzi WW. 2007. Iron Status Is an Important Cause of Anemia in HIV-Infected Tanzanian Women but Is Not Related to Accelerated HIV Disease Progression Journal of Nutrition **137**, 2317–2323.

Massawe, NS. 2002. Anaemia in women of reproductive age in Tanzania. East African Medical Journal **79(9)**, 461-6.

Mata-Marín JA, Gaytán-Martínez JE, Martínez-Martínez ER, Arroyo-Anduiza CI Fuentes-Allen JL. 2010. Casarrubias-Ramirez M. Risk factors and correlates for anemia in HIV treatment-naïve infected patients: a cross-sectional analytical study BMC Research Notes **3**, 230.

McCord JM. 2004. Iron, Free Radicals, and Oxidative Injury. Journal of Nutrition **134**, 3171S–3172S.

McDermid JM, Jaye A, Schim van der Loeff MF, Todd J, Bates C, Austin S, Jeffries D, Awasana AA, Whittlex AA, Prentice A. 2007. Elevated iron status strongly predicts mortality in West African adults with HIV infection. Journal of Acquired Immune Deficiency Syndromes **46**, 498-507.

Millogo A, Sawadogo AB, Sawadogo AP, Lankoandé D. 2002. Neuropathies périphériques révélatrices de l'infection par le VIH au Centre hospitalier de Bobo-Dioulasso (Burkina Faso). Bulletin de la Société de Pathologie Exotique **95(1)**, 27-30.

Mobarak YM. 2011. The *in vivo* and *in vitro* Developmental Toxicity and Teratogenicity of the Anti-AIDS Drugs (Human and Experimental Animal Studies). Trends Applied Science Research **6**, 208-236.

Moh R, Danel C, Sorho S, Sauvageot D, Anzian A, Minga A, Gomis OB, Konga C, Inwoley A, Gabillard D. 2005. Haematological changes in adults receiving a zidovudine-containing HAART regimen in combination with cotrimoxazole in Cote d'Ivoire. Antiviral Therapy **10**, 615-624.

Ministère de la Santé et de l'Hygiène Publique (MSHP). 2009. Rapport Annuel VIH/sida du secteur santé en Cote d'Ivoire 2007-2008.

Nacoulma EWC, Some Y, Tieno H, Diallo I, Zoungrana A, Bougnounou R, Ouédraogo C, Drabo J, Guiard-Schmid JB. 2007. Évolution des paramètres hématologiques au cours du traitement antirétroviral chez les patients infectés par le VIH au Burkina Faso. Bulletin de la Société de Pathologie Exotique **100 (4),** 271-274.

Ngondi, J. L., S.H.L. Etame, M. Fonkoua, H. Yangoua and J. Oben, 2007. Lipid Profile of Infected Patients Treated with Highly Active Antiretroviral Therapy in Cameroon. Journal of Medical Sciences **7**, 670-673.

Nguemaim NF, Mbuagbaw J, Nkoa T, Teto G, Njitchouang GR, Pouomogne DJ, Same-Ekobo A, Asonganyi T. 2010. Changes in Lipid Profiles in Two Groups of HIV-1 Infected Patients in Cameroon on Two Treatment Regimens with Either Efavirenz or Nevirapine, in Association with Reverse Transcriptase Inhibitors. Journal of Medical Sciences **10**, 25-33.

Omoregie R, Omokaro EU, Palmer O, Ogefere HO, Egbeobauwaye A, Adeghe JE, Osakue SI, Ihemeje V. 2009: Prevalence of anaemia among HIVinfected patients in Benin City, Nigeria. Tanzania Journal of Health Research **11**, 1-4.

ONUSIDA, 2009. Directives pour l'élaboration d'indicateurs de base.

Patwardhan MS, Golwilkar AS, Abhyankar JR, Atre MC. 2002. Haematological Profile of HIV positive patients. Indian Journal of *Pathology and* Microbiology **45**, 147-50.

Potdar PD, Daswani BS, Rane NJ. 2011. Drug Resistance Mutations to Protease and Reverse Transcriptase Inhibitors in Treatment Naive HIV-1 Clade C Infected Individuals from Mumbai, International Journal Virology **7**, 13-23.

Russell EC, Charalambous S, Pemba L, Churchyard GJ, Grant AD, Fielding K. 2010. Low haemoglobin predicts early mortality among adults starting antiretroviral therapy in an HIV care programme in South Africa: a cohort study. BMC Public Health **10**,433.

Semba RD, Shah N, Strathdee SA, Vlahov D. 2002. High prevalence of iron deficiency and anemia among female injection drug users with and without HIV infection. Journal of Acquired Immune Deficiency Syndromes **29**, 142–144.

Société de Nutrition et de Diététique de Langue Française (SNDLF). 2001. Anémies nutritionnelles Cahier de Nutrition et de Diététique **36**, 76-81.

Statsoft. 2005. Statistica (Data Analysis Software System). Version 7.1 available at www. Statsoft.com. (accessed 2 february 2006).

Totin D, Ndugwa C, Mmiro F, Perry RT, Jackson JB, Semba RD. 2002. Iron Deficiency Anemia Is Highly Prevalent among Human Immunodeficiency Virus–Infected and Uninfected Infants in Uganda Journal of Nutrition **132**, 423–429.

Trumbo P, Yates AA, Schlicker S, Poos M. 2001. Dietary reference intakes: vitamin A, vitamin K, arsenic, boron, chromium, copper, iodine, iron, manganese, molybdenum, nickel, silicon, vanadium, and zinc. Journal American Dietetic Association **101**, 294–301.

UNICEF/UNU/WHO. 2001. Iron deficiency anemia: assessment, prevention, and control. WHO/NHD/01.3 Geneva, Switzerland: WHO. A guide for programme managers.

Vernet M, Corberand J, David V, Deugnier Y, Frey J, Giraudet P, J-C. Renversez and G. Sebahoun, 2001. Algorithmes de prescription recommandés pour le diagnostic d'un déficit et d'une surcharge en fer. Annales de Biologie Clinique (Paris) **59**, 149-55.

Weiss G, Goodnough LT. 2005. Anemia of chronic disease. New England Journal of Medecine **352(10)**, 1011-23.

Wisaksana R, Sumantri R, Indrati AR, Zwitser A, Jusuf H, Mast QD, Crevel RV, Ven AVD. 2011. Anemia and iron homeostasis in a cohort of HIVinfected patients in Indonesia. BMC Infectious Diseases **11**, 213.