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Research Article

Attitude of leukocyte and thrombocyte parameters during HIV infection among women of childbearing age in the city of Abidjan

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Abstract: The main action of the human immunodeficiency virus (HIV) in the body is to disrupt the immune system by infecting CD4 T cells. This action has dramatic consequences, especially in women of childbearing age. To highlight this state of affairs, a study to evaluate and characterize possible changes in leukocyte and thrombocyte parameters in women of childbearing age in the city of Abidjan (Côte d'Ivoire) was conducted. In fact, 180 women of childbearing age were recruited into a specialized centre for the management of HIV-positive (CIRBA) based on inclusion and exclusion criteria. Blood samples were taken from each of these women to perform their serology and to measure the leukocyte and thrombocyte parameters. The results of these investigations showed that the average values of the thrombocytes in the infected women were altered contrary to the leukocyte parameters. In addition, HIV and antiretroviral therapy (ART) caused an alteration of all the parameters studied. Indeed, a significant difference was observed between them at the level of leukopenia (46.7% vs 30%), neutrophilia (8.3 % vs 1.7%), lymphopenia (20 % vs 3.3%) and low CD4 count (43,3% vs 13,3%), where the proportions were higher in women without ART treatment than in women receiving ARV therapy. This significant difference ($p < 0.05$) was also observed in lymphocytosis (11.7 % vs 36.7 %), thrombocytopenia (0% vs 3.3%) and elevated CD4 count (15% vs 33.3%). At that level, the proportions of elevated lymphocyte values, CD4 lymphocytes, and decreased thrombocyte values decreased in untreated women compared to women on treatment. This study found that non-pregnant women without antiretroviral therapy were most exposed to leukopenia, leukocytosis, neutrophilia, eosinophilia, lymphopenia, and thrombocytosis.

Keywords: HIV-infection, ART, leukocyte, thrombocyte, Côte d'Ivoire.

INTRODUCTION

The immune system of an organism is a biological system consisting to coordinate a set of elements of recognition and defence that discriminates against the "self" of "no self" (De Paoli *et al.*,, 1984). The main effectors of this system are the immune cells called white blood cells produced by stem cells in the red bone marrow (Kohler, 2011). By their function of phagocytosis of bodies or cells such as pathogens (fungi, viruses, bacteria, parasites) and some "foreign" particles or molecules (some toxin), leucocytes have a primary and essential role in the maintenance of the structure and homeostatic functions of tissues (Handin, 2003). In addition, thrombocytes also contribute to homeostasis by their haemostatic function (Rex *et al.*,, 2009).

However, some factors such as infectious and inflammatory syndromes cause immunodeficiency by their harmful actions on the actors of immunity, especially in women of childbearing age (Meda *et al.*,, 2000; Franco et Le Van**,** 2013). These alteration factors include the Human Immunodeficiency Virus (HIV) (Kelley *et al.*,, 2007). Indeed, the Human Immunodeficiency Virus (HIV) disrupts the immune system by infecting CD4 T cells. These cells are the "coordinators" of the immune response and play an exclusively central role in maintaining the physiological integrity of the cells of the body. As a result, a HIV infection degrades the quality and quantity of blood cells. Moreover, this viral infection is widespread in the Africa, particularly in Côte d'Ivoire. In 2010, according to WHO, the national prevalence was 3.9% and seroprevalence in women of childbearing age was 6.4%. Indeed, according to the evaluation of the impact of HIV in the population in Côte d'Ivoire, this prevalence of 6.4 recorded in 2010 decrease to 3.6% in 2018 (CIPHIA, 2018).

Most of the research on the influence of HIV in other developing countries, as well as in Côte d'Ivoire,

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is mainly related to erythrocyte parameters and biological monitoring of patients living with HIV under ARV treatment (Carré *et al.*,, 2003; Sakandé *et al.*,, 2004; Loko *et al.*,, 2005; Kamagaté *et al.*,, 2012a and b; Bléyéré *et al.*,, 2013a, b, c, d and e; Kone *et al.*,, 2019). As well as the effect of certain plants on the strengthening of immunity (Barry, 2014).

Very few scientific studies have revealed the behaviour of leukocyte and thrombocyte parameters during HIV infection in women of childbearing age with or without antiretroviral therapy. Therefore, the general objective of this work was to study and characterize possible changes in leukocyte and thrombocyte parameters in women of childbearing age in the city of Abidjan.

MATERIAL AND METHODS

Site and study population

This study was conducted from 21 October 2009 to 21 December 2010 in the Integrated Research Bio-clinical Centre in Abidjan (CIRBA). It required the voluntary recruitment of 180 women aged 18 to 45 years. Indeed, after information and consent, women who did not have complications of arterial hypertension, diabetes, rheumatoid arthritis were included. In contrast, recently transfused women and women suffering from gynaecological and digestive diseases were excluded. The population's study consisted to recruit:

- 60 healthy non-pregnant women;
- 60 non-pregnant women living with HIV without antiretroviral therapy;
- 60 non-pregnant women living with HIV on antiretroviral therapy.

Blood Samples and Assays of Biological Parameters

In each woman recruited, the blood is taken under aseptic conditions into sterile blood tubes with anticoagulant (EDTA) and in dry tubes of 5 ml each, by venepuncture at the crease of the elbow fasting in the morning. The samples collected on EDTA tubes made it possible to immediately determine the hematological and immunological parameters (CD4 level) with respectively an automatic hematological analyzer Sysmex Xt 2000i and Facscalibur. The samples contained in the dry tubes are centrifuged at 3000 turns for 5 minutes. Fifty (50 µl) microliters of the serum obtained are taken for immediate determination of HIV serology using the Determine and Genie 2 tests.

For HIV serology, the method most used is the successive use of 2 tests. Once the first test (Determine) is positive, the discrimination test (Gen II HIV-1 / HIV-2) is used to determine the type of HIV.

Statistical exploitation of biological parameters

To evaluate the behaviour of leukocyte and thrombocyte parameters during HIV infection in women of childbearing age, with or without treatment, we used a factor analysis of two-factor variances (Anova) (HIV and antiretroviral therapy). The analysis evaluated their dynamics during HIV infection in HIV-infected controls without ARV treatment, and also revealed the possible influence of antiretroviral therapy on the biological parameters of both selected women's groups. Statistical treatments have associated Newman-Keuls' post hoc multivariate test with the post-hoc multivariate test to specify the likely significantly different groups of women with the Statsoft Statistica computer program version Windows 7.1 (Statsoft, 2005).

The different observed proportions of leukocyte and thrombocyte parameters were compared by the test of likelihood G or test log likehood ratio with the Windows version R.2.0.1 software (Ihaka and Gentleman, 1996). The comparison of proportion was made between women of childbearing age constituting controls and women of childbearing age. It was also made between women of childbearing age who were HIV-positive on antiretroviral therapy and those of naive antiretroviral treatment.'

For all statistical operations, a threshold value of probability p less than 0,05 was defined for the significance of the tests.

RESULTS

Characteristics of the population

The general characteristics of the study population are presented in Table 1. The selected women had an average age of 34.7 ± 0.5 years. The body mass index, gestity, parity and the intergenic space had respective mean values of 24.7 ± 0.4 $kg.m^{-2}$; 2.04 ± 0.2; 0.7 ± 0.04; 17.4 ± 1.4 months (Table 1). In this same table, our study selected a low proportion (3.3%) of adolescents compared to subjects whose age is physiologically normal (96.7 %). The body mass index was abnormal (underweight and overweight) in 45%. For our investigations, women were more multi-gest (83.3%), multiparous (52.2%) and more subjects with less than 36 months between pregnancies (68.9%). In addition, women were well educated and included brides, singles and some in a cohabiting relationship.

Among the 120 HIV-positive women, 119 were infected with HIV-1 and one (1) was HIV-1 and 2 positive.

Table 1. General characteristics and types of HIV in the study population.

General Characteristics	Women of childbearing age (N = 180) n %	M±ESM
Age (years)	**34.7 ± 0.5**	
15 - 19	6	3.3
20 - 45	174	96.7
Body mass index (kg.m^{-2})	**24.7 ± 0.4**	
Skinny (< 18,5)	23	12.8
Normal (18,5 – 26)	99	55
Obesity (> 26)	58	32.2
Gravidity	**2.04 ± 0.2**	
Primigravidae	30	16.7
Multigravidae	150	83.3
Parity	**0.7 ± 0.04**	
Nulliparous	56	31.1
Primiparous	0	16.7
Multiparous	94	52.2
Space between births (Months)	**17.4 ± 1.4**	
< 36	124	68.9
> 36	56	31.1
Matrimonial status		
Married	48	26.7
Single	67	37.2
Concubinage	65	36.1
Widowed	0	0
Education attainment		
Uneducated	33	18.3
Primary school	33	18.3
Secondary school	63	35
Superior	51	28.3
Types of HIV		
HIV-1	119	66.1
HIV-2	0	0
HIV-1 et 2	1	0.6

N: Total number of all non-pregnant women; n: Subjects number observed in each group is in brackets; M ± ESM: Average ± Standard error on the average.

HIV and leukocyte and thrombocyte parameters in women of childbearing age

The results of the study showed that the average values of the leukocyte and thrombocyte parameters with infected women of childbearing age, on the one hand, and uninfected one on the other hand, were normal (Table 2). According to the comparison of the means of these biological parameters, only the level of thrombocytes revealed a significant difference between infected and non-infected women ($p < 0.05$). This thrombocyte parameter was significantly low with infected women compared to uninfected one. On the other hand, all the leukocyte parameters, namely leukocytes, lymphocytes, monocytes and polynuclear cells (neutrophils; eosinophils; basophils) presented statistically equal mean values in both infected and uninfected women.

The results indicated in Table 3 reported the proportions of the leukocyte and thrombocyte parameters in non-pregnant control and HIV-infected women. Leukopenia and leukocytosis were higher in infected non-pregnant women than in control women. Similarly, neutropenia, eosinophilia, monocytosis, and thrombocytopenia were higher with non-pregnant women who were infected compared to the control one. Conversely, the prevalence of neutropenia, basophilia, lymphocytosis and thrombocytosis was lower in infected women compared to control one. The differences observed were not statistically significant ($p > 0.05$). However, the proportion of neutrophilia was significantly higher in infected women (5%) than in control one (0%).

Table 2: HIV and behaviour of leukocyte and thrombocyte parameters in women of childbearing age.

Leukocyte and thrombocyte parameters	HIV negative women of childbearing age (control, N = 60)	HIV positive women of childbearing age women (N = 120)	p
Leukocyte (10^9/l)	5 ± 0.2	4.6 ± 0.2	0.06 (NS)
P. Neutrophils (%)	44.1 ± 1.6	43.5 ± 1.2	0.9 (NS)
P. Eosinophils (%)	3.1 ± 0.4	4.1 ± 0.4	0.8 (NS)
P. Basophils (%)	0.5 ± 0.04	0.4 ± 0.02	0.8 (NS)
Monocytes (%)	9.4 ± 0.4	11.2 ± 0.4	0.06 (NS)
Lymphocytes (%)	42.4 ± 1.5	40.8 ± 1.2	0.08 (NS)
Thrombocytes (10^9/l)	269 ± 15.2	258.7 ± 9.8	0.04 (S)

N: total number of each group of women; P: Polynuclear; S: Statistically different for p value < 0,05; NS: Not statistically significant for p value > 0.05.

Table 3: HIV and proportions of leukocyte and thrombocyte parameters in women of childbearing age

Leukocyte and thrombocyte parameters	HIV negative women of childbearing age (control, N = 60)	HIV positive women of childbearing age (N = 120)	p
	n (%)	n (%)	
Leucocyte (10^9/l)			
Leukopenia (< 4)	22 (36.7)	46 (38.3)	0.8 (NS)
Normal (4-10)	38 (63.3)	78 (60.8)	0.8 (NS)
Leukocytosis (> 10)	0 (0)	1 (0.8)	0.3 (NS)
Neutrophils (%)			
Neutropenia (< 45)	36 (60)	70 (58.3)	0.9 (NS)
Normal (45-70)	24 (40)	44 (36.7)	0.7 (NS)
Neutrophilia (> 70)	0 (0)	6 (5)	**0.01 (S)**
Eosinophils (%)			
Normal (0-4)	44 (73.3)	81 (67.5)	0.6 (NS)
Eosinophilia (> 4)	16 (26.7)	39 (32.5)	0.4 (NS)
Basophils (%)			
Normal (0-0.5)	43 (71.7)	88 (73.3)	0.9 (NS)
Basophilia (> 0.5)	17 (28.3)	32 (26.7)	0.8 (NS)
Monocytes (%)			
Monocytopenia (< 3)	0 (0)	0 (0)	
Normal (3-10)	38 (63.3)	59 (49.2)	0.2 (NS)
Monocytosis (> 10)	22 (36.7)	61 (50.8)	0.1 (NS)
Lymphocytes (%)			
Lymphopenia (< 25)	5 (8.3)	14 (11.7)	0.5 (NS)
Normal (25-50)	39 (65)	77 (64.2)	0.9 (NS)
Lymphocytosis (> 50)	16 (26.7)	26 (21.7)	0.5 (NS)
Thrombocytes (10^9/ l)			
Thrombocytopenia (< 150)	0 (0)	2 (17)	0.1 (NS)
Normal (150-400)	53 (88.3)	105 (87.5)	0.9 (NS)
Thrombocytosis (> 400)	7 (11.7)	13 (10.8)	0.8 (NS)

N: total number of each group of women; n: Subjects number observed in each group is in brackets; %: Proportion indicated in each subjects group; S: Statistically different for p value < 0.05; NS: Not statistically significant for p value > 0.05.

Antiretroviral therapy and leukocyte and thrombocyte parameters in women of childbearing age

The comparison of the various leukocyte and thrombocyte parameters of uninfected women treated and without treatment indicated that the infected women of childbearing age who are under antiretroviral treatment, presented a statistically lower CD4 value (275.5 ± 34.3) (p < 0.01) than those without treatment (463.1 ± 33.9).

The other leukocyte parameters showed no significant difference (p > 0.05) between the two groups of women (Table 4).

The proportions of the leukocyte and thrombocyte parameters in the two groups of women of childbearing age infected with HIV are reported in Table 5. The data indicated leukocytosis (p = 0.1), neutropenia (p = 0.1), eosinophilia (p = 0.8), basophilia (p = 0.4), monocytopenia, monocytosis (p = 0.9) and thrombocytosis (p = 0.03) statistically equal in these two groups of women. However, a significant difference was observed between them in terms of leukopenia (p = 0.04), neutrophilia (p = 0.03), lymphopenia (p = 0.0003) and low CD4 count. (p = 0.00004) where the proportions were higher in women without ARV treatment than in those who are on ARV treatment. This significant difference (p < 0.05) was also observed in terms of lymphocytosis (p = 0.0002),

thrombocytopenia (p = 0.03) and the high CD4 count (p = 0.008). At this level, the proportions of high values of lymphocytes and CD4, and decrease values of thrombocytes decreased in women without treatment compared to one under treatment.

Table 4: Antiretroviral treatment and variation of leukocyte and thrombocyte parameters in women of childbearing age.

Leukocyte and thrombocyte parameters	HIV positive women naïve ART (N = 60)	HIV positive women with ART (N= 60)	p
Leucocyte (10^9/l)	4.4 ± 0.3	4.8 ± 0.2	0.07 (NS)
P. Neutrophils (%)	47.9 ± 1.8	39.1 ± 1.5	0.06 (NS)
P. Eosinophils (%)	4 ± 0.5	4.3 ± 0.6	0.8 (NS)
P. Basophils (%)	0.4 ± 0.04	0.4 ± 0.03	0.9 (NS)
Monocytes (%)	11.3 ± 0.6	11.1 ± 0.6	0.9 (NS)
Lymphocytes (%)	36.4 ± 1.7	45.1 ± 1.5	0.06 (NS)
Thrombocytes (10^9/l)	270 ± 15.3	247.3 ± 12.3	0.03 (NS)
CD4+ (Cell/mm^3)	275.5 ± 34.3	463.1 ± 33.9	**0.01 (S)**

N: total number of each group of women; ART: antiretroviral therapy; P: Polynuclear; S: Statistically different for p value < 0.05; NS: Not statistically significant for p value > 0.05.

Table 5: Antiretroviral treatment and proportions of leukocyte and thrombocyte parameters in women of childbearing age

Leukocyte and thrombocyte parameters	HIV positive women naïve ART (N = 60)	HIV positive women with ART (N= 60)	p
	n (%)	n (%)	
Leucocyte (10^9/l)			
Leukopenia (< 4)	28 (46.7)	18 (30)	**0.04 (S)**
Normal (4-10)	31 (51.7)	42 (70)	0.10 (NS)
Leukocytosis (> 10)	1 (1,7)	0 (0)	0,1 (NS)
Neutrophils (%)			
Neutropenia (< 45)	30 (50)	40 (66.7)	0.1 (NS)
Normal (45-70)	25 (41.7)	19 (31.7)	0.2 (NS)
Neutrophilia (> 70)	5 (8.3)	1 (1.7)	**0.03 (S)**
Eosinophils (%)			
Normal (0-4)	40 (66.7)	41 (68.3)	0.9 (NS)
Eosinophilia (> 4)	20 (33.3)	19 (31.7)	0.8 (NS)
Basophils (%)			
Normal (0-0.5)	46 (76.7)	42 (70)	0.6 (NS)
Basophilia (> 0.5)	14 (23.3)	18 (30)	0.4 (NS)
Monocytes (%)			
Monocytopenia (< 3)	0 (0)	0 (0)	-
Normal (3-10)	29 (48.3)	30 (50)	0.9 (NS)
Monocytosis (> 10)	31 (51.7)	30 (50)	0.9 (NS)
Lymphocytes (%)			
Lymphopenia (< 25)	12 (20)	2 (3.3)	**0.0003 (S)**
Normal (25-50)	41 (68.3)	36 (60)	0.5 (NS)
Lymphocytosis (> 50)	7 (11.7)	22 (36.7)	**0.0002 (S)**
Thrombocytes (10^9/ l)			
Thrombocytopenia (< 150)	0 (0)	2 (3.3)	**0.03 (S)**
Normal (150-400)	52 (86.7)	53 (88.3)	0.9 (NS)
Thrombocytosis (> 400)	8 (13.3)	5 (8.3)	0.3 (NS)
CD4 (Cell/mm^3)			
Low (< 200)	26 (43.3)	8 (13.3)	**0.00004 (S)**
Normal (200-499)	25 (41.7)	32 (63.3)	0.03 (NS)
High (≥500)	9 (15)	20 (33.3)	**0.008 (S)**

N: total number of each group of women; n: Subjects number observed in each group is in brackets; %: Proportion indicated in each subjects group; S: Statistically different for p value < 0.05; NS: Not statistically significant for p value > 0.05.

DISCUSSION

The study conducted in the population of women of childbearing age in the city of Abidjan showed the general alteration of the leukocyte and thrombocyte parameters during HIV infection. This alteration mainly concerned the average thrombocyte values with infected women. On the other hand, the leukocyte parameters remained almost stable. However, the average CD4 count dropped sharply among those who did not receive antiretroviral therapy. In addition, in 5% of women of childbearing age, HIV causes neutrophilia. This result differs to the one of Loua *et al.*, (2011) in Mali who found neutropenia in 15.7% of people living with HIV. However, the neutropenia observed in 58.3% of the women infected in our study was not significantly different from the one of uninfected women (60%). This prevalence of neutropenia observed in our study is similar to what had been observed by Duran (2005). This author found between 60 and 75% of cases of neutropenia during their investigation in seropositive people.

In this population, antiretroviral therapy has significantly influenced the prevalence of leukopenia, neutrophilia, lymphopenia, lymphocytosis and thrombocytopenia, also modifying the CD4 count. This change in CD4 count has been demonstrated in study of Lozès *et al.,* (2012) and Ukibe *et al.,* (2012) in Benin and Nigeria respectively. Indeed, the entry of HIV into the body causes an inflammatory process which immediately mobilizes the neutrophils, hence the neutrophilia observed through infected women without treatment. Lymphopenia and the decrease of CD4 count could be explained by the infection of CD4 T cells by HIV, followed by their death when the newly formed viruses leave. According to Delabesse *et al.,* (2010), lymphopenia is a constitutive element of AIDS with a quantitative and qualitative deficit of CD4 T lymphocytes. This dramatic reduction in lymphocyte levels is believed to be the cause of leukopenia seen through infected women without treatment. These different haematological anomalies observed in our results are similar to those found by Kelley *et al.,* (2007) who showed that 35.26% of people with HIV have leukopenia, 30% lymphopenia and 74 % thrombocytopenia during the first -infection. All these pathologies induced by HIV infection, as observed by Kelley *et al.,* (2007) could be restored by antiretroviral therapy which has the role of blocking the replication cycle of the virus and therefore reducing the viral load as indicated Thaczuk *et al.,* (2011). Low prevalence of these above-mentioned pathologies has been observed with women infected with antiretroviral therapy.

Indeed, taken simultaneously, the impact of HIV and antiretroviral therapy, unlike the previous results, caused neutropenia, lymphocytosis and mild thrombocytopenia in women infected with ARV therapy. These results are similar to those of Talom *et al.,* (2005) and Kazadi *et al.,* (2009), who also observed neutropenia and thrombocytopenia during antiretroviral therapy with infected people but with different proportions.

All of the biological parameters studied underwent significant changes in all groups of women. These changes are mainly observed in neutropenia, basophilia, lymphocytosis and thrombocytopenia.

Furthermore, the interaction between HIV and ARV strongly influences CD4 levels. These CD4 counts were lower with infected women without ARV treatment than non-pregnant women on antiretroviral therapy. Antiretroviral therapy is therefore believed to be responsible for restoring the immune system following immunodeficiency caused by HIV by preventing the destruction of T-CD4 lymphocytes, the main actors in immune reactions (Chirouze and Hoen, 2006).

Considering both groups of women studied, it is non-pregnant women infected without antiretroviral therapy who are the most exposed to leukopenia, leukocytosis, neutrophilia, eosinophilia, lymphopenia and thrombocytosis. This could be explained by the weakened immune system to HIV infection.

CONCUSION

In our study, different haematological anomalies were observed. Concerning the thrombocyte line, there is a predominance of thrombocytopenia due to infection.

An immune deficiency characterized by a decrease of CD4 T cells has been observed with women of childbearing age infected without treatment.

Among all of groups of women studied, it is women of childbearing age infected without ARV treatment who are the most exposed to these pathologies. Antiretroviral therapy therefore allows these leukocyte and thrombocyte parameters to evolve in the direction of improving their values. However, it is responsible for neutropenia and lymphocytosis. This work indicates the need of adequate biological monitoring for better care of women of reproductive age infected with HIV in Côte d'Ivoire.

COMPETING OF INTEREST

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

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.

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