

## POSSIBLE IMPACT OF CO-INFECTIONS OF TUBERCULOSIS AND MALARIA ON THE CD4<sup>+</sup> CELL COUNTS OF HIV PATIENTS IN NIGERIA

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### Key words

HIV, Co-infection, Tuberculosis, Malaria, CD4<sup>+</sup> cell count

### Abstract

**Background:** This study focused on evaluating the possible impact of co-infections of tuberculosis and malaria on the CD4<sup>+</sup> cell counts in HIV infected subjects.

**Methods:** This is a cross sectional study. The subjects were drawn from three hospitals and a blood bank in Lagos State. After due consent, blood samples were obtained from 69 subjects with single infections (HIV, TB, and Malaria), 34 subjects with multiple infections (HIV/Malaria, HIV/TB, Malaria/TB, HIV/TB/Malaria) and 24 blood donors (controls). The CD4<sup>+</sup> cell counts of all the 127 blood samples were estimated using a FACS count.

**Results:** Data obtained were analysed and a comparison of the results showed that the median CD4<sup>+</sup> counts in all groups of subjects with HIV infections (whether single or co-infection) were similar and significantly lower than the median counts for the healthy control group as well as groups without HIV infection (malaria, TB and malaria/TB).

**Conclusion:** Overall data further confirmed the progressive depletion of CD4<sup>+</sup> cells in HIV infection while co-infections with TB and malaria did not have any impact on the CD4<sup>+</sup> cells of HIV infected subjects. A larger prospective study is needed.

### Mots clés

VIH, la Co-Infection, la Tuberculose, le Paludisme, le compte de cellule, CD4<sup>+</sup>

### Résumé

**Fond:** Cette étude a été consacrée à l'évaluation de l'impact possible de co-infections de tuberculose et le paludisme sur les comptes de cellule CD4<sup>+</sup> des sujets infectés du VIH.

**Méthode:** Ceci est une étude transversale. Les sujets ont été choisis de trois différents hôpitaux et une banque du sang dans l'Etat de Lagos. Après le consentement nécessaire, les échantillons de sang ont été obtenus de 69 sujets avec les mono-infections (VIH, TB, et le Paludisme), 34 sujets avec les infections multiples (le VIH/PALUDISME, LE VIH/TB, LE Paludisme/TB, VIH/TB/le Paludisme) et 24 donneurs de sang (les contrôles). les comptes de cellule CD4<sup>+</sup> de tous les 127 échantillons de sang ont été estimés utilisant une compte FACS.

**Résultats:** les données obtenues ont été analysées et une comparaison des résultats a démontré que le médian des comptes CD4<sup>+</sup> dans tous les groupes de sujets avec les infections de VIH (soit mono ou co-infection) étaient similaires et significativement plus bas que les comptes médianes pour le groupe de contrôle sain de même que les groupes sans l'infection de VIH (le paludisme, TB et le paludisme/TB).

**Conclusion:** les données générales ont confirmé le plus l'épuisement progressif des cellules CD4<sup>+</sup> dans l'infection de VIH pendant que les co-infections avec TB et le paludisme n'ont pas eu aucun impact sur les cellules CD4<sup>+</sup> des sujets infectés de VIH. Une plus profonde étude sera nécessaire.

## Introduction

Human Immunodeficiency Virus (HIV), infection has been associated with a reduction in the absolute numbers of CD4<sup>+</sup> T-lymphocytes.<sup>1</sup> Thus one of the best measurements for monitoring disease progression in HIV-infected individuals has been by the determination of the CD4<sup>+</sup> T- lymphocyte count.<sup>2</sup> The CD4<sup>+</sup> cell counts have also been used for the staging of HIV infections as well as monitoring of response to antiretroviral therapy. A profile of opportunistic infections has been associated with HIV/AIDS.<sup>3</sup> Equally concurrent infections of prevalent endemic diseases with HIV / AIDS have been reported.<sup>4</sup>

Findings from several studies suggest that active tuberculosis may accelerate HIV-induced immunologic deterioration. The active tuberculosis is associated with transient CD4<sup>+</sup> T-lymphocyte depression.<sup>5, 6</sup> Also, tuberculosis results in immune stimulation and the increased production of cytokines such as tumour necrosis factor<sup>7 - 9</sup> which increase HIV replication in vitro.<sup>10, 11</sup> Moreover, HIV-infected patients with tuberculosis appeared to have a higher risk of opportunistic infections and death than HIV-infected patients with similar CD4<sup>+</sup> T- cell counts but without tuberculosis.<sup>12</sup>

In a cohort of HIV-infected individuals, the incidence of clinical malaria was found to have increased with decreasing CD4 cell counts.<sup>13</sup> In a population- based cohort, HIV- infected individuals had a significantly higher risk of having parasitaemia and developing clinical malaria than HIV- negative individuals. This risk tended to increase with decreasing CD4 cell counts.<sup>14</sup> Considering the effect of malaria on HIV infection, because malaria induces CD4 cell activation<sup>15</sup> and increases the concentration of proinflammatory cytokines in plasma<sup>16</sup> it is expected to enhance HIV replication<sup>17</sup> and thus accelerate HIV disease progression.<sup>18</sup> Since the CD4<sup>+</sup> cell counts has been an important surrogate marker for HIV prognosis, attempts were made in this study to examine any possible impact of opportunistic or concurrent infections with HIV/AIDS on the CD4<sup>+</sup> cell counts. This pilot study focused on Tuberculosis (TB), as the most prevalent opportunistic infection and Malaria, as the most prevalent endemic infection in Nigeria.

## Materials and Methods

The study was cross-sectional in design and it comprised of a population of 127 adult subjects of both sexes aged 18 years and above. The distribution of the disease conditions of the subjects were 21 (HIV+ve), 24 (TB+ve), 24 (Malaria +ve), 10 (HIV + Malaria+ve), 11(HIV + TB+ve), 8 (Malaria + TB+ve) and 5 (HIV + TB + Malaria+ve) and 24(Blood

donors) who tested negative for HIV, TB and Malaria and thus regarded as apparently healthy. This last group served as controls. A total of 69 subjects had single infections while 34 subjects had multiple infections.

The subjects were drawn from patients who presented between September to November 2001 at the Lagos University Teaching Hospital, the Mainland Hospital Yaba, the Staff Clinic of the Nigerian Institute of Medical Research and the blood bank of the National Orthopedic Hospital Lagos. The disease conditions of these subjects had earlier been diagnosed in these hospitals.

After due counseling and informed consent obtained, 10ml of blood as well as sputum samples were collected from each subject. The blood samples were screened at the NIMR laboratory to reconfirm the HIV and Malaria status of the patients. The sputum samples were screened for AFB to reconfirm the TB status. The CD4<sup>+</sup> cell counts were estimated from aliquots of each of the blood samples using the FACS count technique. All data generated were collated and analysed using Epi-INFO Version 6.04.

## Results

Data showed that the study population comprised almost as many males (51%) as females (49%). The median CD4<sup>+</sup> cell counts estimated for the subjects with single infections were 386 cells/ $\mu$ l, 787 cells/ $\mu$ l and 737 cells/ $\mu$ l for the HIV, TB and Malaria subjects respectively (Table 1). The median count recorded for the apparently healthy subjects (control) was 789 cells/ $\mu$ l.

The median counts recorded for the TB and Malaria patients were not significantly different from the median count recorded for the healthy controls ( $p > 0.05$ ). Furthermore, the median count recorded for the HIV subjects was lower than for the TB and Malaria subjects and significantly lower than for the healthy controls ( $p < 0.001$ ).

For the group of subjects with co-infections, the lowest median CD4<sup>+</sup> cell count of 182 cells/ $\mu$ l was recorded for subjects co-infected with HIV, TB and Malaria (Table 2). A median count of 211 cells/ $\mu$ l was recorded for subjects with HIV and Malaria co-infections while median counts of 268 cells/ $\mu$ l and 1007 cells/ $\mu$ l were respectively recorded for subjects with HIV/TB and Malaria/TB co-infections. The median counts for each of the three groups that had co-infections with HIV, were very much lower than the median counts obtained for the Malaria/TB multiple infection. In fact, their median CD4 cell counts were not statistically different from that of HIV single infection. While the median count recorded for the Malaria/TB subjects was found to be significantly higher than HIV single infection ( $p < 0.001$ ).

Table 1: Impact of single disease conditions of CD4<sup>+</sup> cell counts (cells/ $\mu$ l)

Single infection	n	Median	Mean	SD	*p value
HIV	21	386.0	426.5	373.2	<0.001
Tuberculosis	24	787.0	875.5	353.8	0.813
Malaria	24	737.0	739.9	219.1	0.14
Apparently healthy	24	789.0	853.0	297.6	

HIV: Human Immunodeficiency Virus

SD: Standard deviation

\*p-values as compared with apparently healthy controls

Table 2: Impact of co-infections with HIV on CD4<sup>+</sup> cells count (cells/ $\mu$ l)

Multiple infections	n	Median	Mean	SD	*p value
HIV and malaria	10	211.0	229.6	202.5	0.130
HIV and tuberculosis	11	268.0	427.2	308.9	0.996b
Malaria and tuberculosis	8	1007.0	1076.8	420.5	<0.001
HIV, malaria and tuberculosis	5	182.0	233.2	147.0	0.271

\*p-values were as compared with HIV single infection

## Discussion

The significant observation in this study was that in the two broad groups comprising of single and co-infections, subjects with HIV infections had more depleted levels of CD4<sup>+</sup> cell counts. Though the CD4<sup>+</sup> cell counts were much more depleted in HIV co-infections than in HIV single infection there was no statistical difference. The level of CD4<sup>+</sup> T-cells in these HIV subjects (either as single or co-infections), were much lower than the reported normal range of 500-1500 cells/ $\mu$ l for the general Nigerian population.<sup>19, 20</sup> This obviously could be adduced to the fact that the HIV infects mainly the CD4<sup>+</sup> cells.<sup>21</sup> The levels of CD4<sup>+</sup> cell counts of subjects with either single infections of TB, Malaria or multiple infections with TB/Malaria were within the normal range reported for the adult Nigerian population. This implies that neither TB nor malaria infections have direct impact on the CD4<sup>+</sup> cells.

However, studies have shown that rates of TB and malaria fever increased with lower CD4<sup>+</sup> cell counts.<sup>14</sup> Reports from Uganda,<sup>22</sup> Kenya,<sup>23</sup> South Africa<sup>24</sup> have shown that rates of tuberculosis and malaria fever increased with lower CD4<sup>+</sup> cell counts. In other words, TB or malaria infections do not result in the depletion of CD4<sup>+</sup> cells but there is an increasing risk of TB and malaria fever infections with advancing HIV immunosuppression. On a population basis, an increased prevalence of malaria and tuberculosis could lead to increased malaria and tuberculosis transmission affecting both HIV positive and negative individuals. The increased risk of these infections in HIV positive subjects could increase the burden on clinical services in areas with high prevalence of HIV-1.<sup>14</sup> The median CD4<sup>+</sup> cell count among the apparently healthy was within the reported normal range for Nigerians as expected.

The overall finding further confirmed that in Nigerian subjects as in other nationals, HIV infection progressively depletes the CD4<sup>+</sup> T-lymphocyte levels

of the infected subject. Also co-infection with TB or Malaria appeared not to have any impact on the level of depletion of CD4<sup>+</sup> T-lymphocyte in the HIV infected subjects as the cell counts were similar to those infected with HIV single infection. However, this is a cross-sectional study with a limited population size. A larger and prospective study will be needed to indeed establish the impact of TB and malaria co-infections on HIV disease progression.

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