The Correlation Between Total White Blood Cell Count, Total Lymphocyte Count, & the CD4+ T-helper Lymphocyte Count among Iraqi HIV Infected Patients

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Abstract:

Study of the correlation between changes in the white blood cell count (WBC), total lymphocyte count (TLC) and the CD4+ T-helper lymphocyte count (CD4), have been carried out during Oct. 2004 through April. 2005 among 35 HIV seropositive Iraqi patients and 30 seronegative individuals. ELISA and Western blot assays were used to detect and confirm the presence of anti-HIV antibodies in the blood samples.

Patients and control groups have been classified into (4) categories according to the obtained results of CD4 count: <200, 200-500, 500-1000, and >1000 cell/cmm. This categorization shows that: none of the HIV seropositives was found to be within the first category; six within the second category (mean CD4 = 383.2 cell/cmm, SD= 92.6) twenty-one within the third category (mean CD4 = 787.9 cell/cmm, SD= 131.6); and eight within the fourth category (mean CD4= 1237.1 cell/cmm, SD= 159.4). While among the control group: none of them was found to be within the first category; only one within the second category (CD4= 464 cell/cmm); twenty-six within the third category (mean CD4 = 745.7 cell/cmm, SD= 116.2); and three within the fourth category (mean CD4= 1333 cell/ cmm, SD= 331).

Results showed that among HIV seropositives a strong significant, positive correlation was found between changes in CD4 and TLC within the second and the third categories, and even when the whole data (uncategorized) were used. While there was a moderate significant positive correlation within the fourth category. The correlation between total WBC and CD4 was found to be insignificant.

Key words: Human Immunodeficiency virus (HIV), white blood cell count, Total lymphocyte count (TLC), CD4+ T-helper lymphocyte count (CD4), Acquired immunodeficiency syndrome (AIDS).

Introduction:

Acquired immunodeficiency syndrome (AIDS) is a severe disease representing the late clinical stage of HIV infection. This disease was recognized for the first time in 1981(1).

CD4+ T-helper lymphocyte (CD4), which are depleted in AIDS patients, are the primary targets of HIV, because of the affinity of the gp120 (a glycoprotein component of the viral envelope) for the CD4 molecules found on the cell surface of these cells (2, 3). The depletion of CD4 is the hallmark of the HIV infection; it is a predictor of an individual at risk for infection with opportunistic pathogens, as well as other complications of HIV infection (4, 5).

According to the laboratory information; patients are categorized according to CD4 count into three groups or categories (>500cell/cmm, 200-500 cell/cmm, and <200 cell/cmm), or according to
the total lymphocyte count (TLC) into (>2000 cell/cmm, 1000-2000 cell/cmm, and <1000 cell/cmm) this system was found prognostically valuable \(^6,^7\).

In Iraq the HIV infection was discovered for the first time in 1986, since that time to the end of 2004, 260 persons were found to be infected with HIV\(^8\).

**Patients & Methods:**

Thirty Five persons who were HIV- antibody seropositive and 30 seronegative control group, from whom 10 ml of peripheral blood sample have been collected. The blood samples have been tested with screening assay (anti-HIV TETRA ELISA; Biotest, Germany); seropositive samples then have been tested with confirmatory assay (HIV blot 2.2 western blot assay; Genelabs diagnostic pte ltd, Singapore).

A total white blood cells (WBC) was counted using Turk solution and Neubaure`s chamber, a differential white blood cell count (DWBC) to determine the percentages of each type of the leukocytes using Lieszman's stain \(^9,^{10}\) were made. The total number of lymphocytes per cubic milliliter (TLC) was calculated using the following formula:

\[
TLC = \frac{\text{Lymphocyte} \times \text{WBC}}{100}
\]

CD4\(^+\) T-helper lymphocytes were counted using the immunoflourescence technique based on the anti-CD4 antibodies according to the manufacturer manual (Sanqiuin, Holland).

The Minitab 11 statistical program was used in testing the significance, calculating the correlation equation and the confidence coefficient between each parameter and the CD4 count. A scattergram showing the correlation also had been drawn using the same program \(^{11}\).

**Results:**

None of the HIV- patients was found to have CD4 count less than 200 cell/cmm, while only 6 of them were found to have CD4 count ranging from 200-499 cell/cmm; table (1) shows the frequencies of patients and controls in different CD4 count categories. The WBC of only 8 patients was found to be less than the normal range; table (2) shows the total white blood cells count in different CD4 count categories and the correlation value \((r)\) between CD4 and WBC count among each category, which demonstrate that no significant correlation was found between these two counts. Total lymphocyte count among HIV patients was found to fall within the range of 700-4048 cell/cmm, and as demonstrated in table (3) there was a significant correlation ranging from strong \((r=0.957)\) to moderate \((r=0.681)\) between changes in this count and CD4 count suggesting that this marker is a good marker to be used as a predictor of disease progression in HIV infected patients when CD4 counting is not available.

In addition to the calculation of correlation value, the obtained data were used to draw scattergrams to show the correlation between changes in CD4 count, changes in TLC and WBC counts.

Figures (1) and (2) show that the correlation between changes in CD4 and WBC counts was strong \((r=0.828)\) and direct when the whole data were used, but this correlation tend to become weak \((r=0.242)\) when the data of patients with CD4 count of <500 cell/cmm were used. Figures (3) and (4) show a strong significant direct correlation between changes in CD4 and TLC counts among patients group when the whole data \((r=0.93)\) and that of patients with CD4 count <500 cell/cmm \((r=0.97)\) were used respectively.

**Table (1):** Frequencies in different CD4 count categories.
<table>
<thead>
<tr>
<th>CD4 T-cell Category (group)</th>
<th>Tested Group</th>
<th>No. (%)</th>
<th>Min. Cell/cmm</th>
<th>Max. Cell/cmm</th>
<th>Mean± SD. Cell/cmm</th>
<th>Elevated No. (%)</th>
<th>Decreased No. (%)</th>
<th>Correlation Value (r)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>1st</strong> (&lt;200 cell/cmm)</td>
<td>Patients</td>
<td>0 (0)</td>
<td>/</td>
<td>/</td>
<td>/</td>
<td>/</td>
<td>/</td>
<td>/</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>0 (0)</td>
<td>/</td>
<td>/</td>
<td>/</td>
<td>/</td>
<td>/</td>
<td>/</td>
</tr>
<tr>
<td><strong>2nd</strong> (200-499 cell/cmm)</td>
<td>Patients</td>
<td>6 (17.1)</td>
<td>244</td>
<td>461</td>
<td>383.2±92.6</td>
<td>0 (0)</td>
<td>4 (66.6)</td>
<td>0.242 W*</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>1 (3.3)</td>
<td>464</td>
<td>464</td>
<td>464</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td>/</td>
</tr>
<tr>
<td><strong>3rd</strong> (500 – 1000 cell/cmm)</td>
<td>Patients</td>
<td>21(60)</td>
<td>510</td>
<td>979</td>
<td>687.9±131.6</td>
<td>0 (0)</td>
<td>4 (19)</td>
<td>0.541 M**</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>26(86.6)</td>
<td>551</td>
<td>924</td>
<td>745.7±116.2</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td>0.03 W</td>
</tr>
<tr>
<td><strong>4th</strong> (&gt;1000 cell/cmm)</td>
<td>Patients</td>
<td>8(22.8)</td>
<td>1008</td>
<td>1428</td>
<td>1237.1±159.4</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td>0.449 W</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>3(10)</td>
<td>1107</td>
<td>1713</td>
<td>1333±331</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td>0.376 W</td>
</tr>
</tbody>
</table>

* Weak, ** Moderate
Table (3): Total lymphocyte count in different CD4 counts categories.

<table>
<thead>
<tr>
<th>CD4 T-cell Category (group)</th>
<th>Tested Group</th>
<th>No. (%</th>
<th>Min. Cell/c mm</th>
<th>Max Cell/c mm</th>
<th>Mean ± SD. Cell/c mm</th>
<th>Elevated No. (%)</th>
<th>Decreased No. (%)</th>
<th>Correlation Value (r)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2nd (200-499 cell/cmm)</td>
<td>Patients</td>
<td>6 (17.1)</td>
<td>700</td>
<td>1235</td>
<td>1003.7±195.2</td>
<td>1 (16.6)</td>
<td>2 (33.3)</td>
<td>0.975 S*</td>
</tr>
<tr>
<td>Control</td>
<td>1 (3.3)</td>
<td>2115</td>
<td>2115</td>
<td>2115</td>
<td>1 (100)</td>
<td>0 (0)</td>
<td>/</td>
<td></td>
</tr>
<tr>
<td>3rd (500 – 1000 cell/cmm)</td>
<td>Patients</td>
<td>21(60)</td>
<td>1134</td>
<td>2400</td>
<td>1703±382.5</td>
<td>7 (33.3)</td>
<td>1 (4)</td>
<td>0.737 S</td>
</tr>
<tr>
<td>Control</td>
<td>26(86.)</td>
<td>1452</td>
<td>3048</td>
<td>2372±523</td>
<td>15 (57.6)</td>
<td>4 (15.3)</td>
<td>0.181 W</td>
<td></td>
</tr>
<tr>
<td>4th (&gt;1000 cell/cmm)</td>
<td>Patients</td>
<td>8(22.8)</td>
<td>2340</td>
<td>4048</td>
<td>3117±636</td>
<td>6 (75)</td>
<td>0 (0)</td>
<td>0.681 M</td>
</tr>
<tr>
<td>Control</td>
<td>3(10)</td>
<td>2944</td>
<td>3496</td>
<td>3232±277</td>
<td>2 (66.6)</td>
<td>0 (0)</td>
<td>0.762S</td>
<td></td>
</tr>
</tbody>
</table>

* Strong

Figure (1): Scatter gram showing the correlation between CD4 and total white blood cells counts among HIV infected patients.

Figure (2): Scatter gram showing the correlation between CD4 and white blood cell counts among HIV infected patients with CD4 count < 500 cell/cmm.
Discussion:

The correlation between progression to AIDS and changes in WBC, TLC counts and other hematological markers was the main target for many researches; the aim of all these researches was to find the best (accurate, cheap, and simply done) predicting markers of disease progression in HIV infected patients so as to determine when to start or to stop antiretroviral therapy in order to avoid the complete dysfunction of the immune system and to avoid opportunistic infections\(^1\)\(^,\)\(^2\). In this study we follow the same target.

The correlation between disease progression and changes in CD4 T-lymphocyte count has been proved by several studies that have been done since the discovery of the disease\(^3\)\(^-\)\(^7\).

In the present study we used the CD4 as a baseline to which the correlation of changes in WBC and TLC has been statistically analyzed and despite the increase or decrease in these markers among different CD4 count groups. According to our results only the TLC was found to have a strong correlation to the changes in CD4 among all CD4 count groups (table 2; Figures 3, and 4). Several studies have suggested the usefulness of TLC to be used to predict the CD4 count in HIV/AIDS patients\(^8\)\(^-\)\(^12\). Our results came in accordance with these suggestions. However, others\(^1\) in their study suggested that TLC is not a good predictor of the CD4 in HIV/AIDS patient population. Nevertheless they also mentioned that if no alternative assay is available to determine the CD4, TLC can be used, taking into account the clinical conditions, and risk factors for developing opportunistic infections in the patients. This status was found in Iraq where CD4 counting technique is not available and total white blood cell (WBC) and differential white blood cell counting are easy to determine using very cheap technique.

References:


8. AIDS Research's and studies center, personal communications.


