**EFFECT OF EXAMINATION STRESS ON BLOOD CELLS PARAMETERS AMONG MEDICAL TECHNOLOGY STUDENTS IN THE UNIVERSITY OF ZAWIA, LIBYA**

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**ABSTRACT**

Medical university examinations are known to cause mental stress. Stress can lead to various changes in the normal functioning of the human body. This study was carried out among student of faculty of medical technology in University of Zawia. This investigation aimed to determine whether academic stress can affect the levels of WBCs, lymphocytes, hemoglobin, and platelets of students. Forty samples were collected from both male and female students aged 18 to 23 years. The collection was carried out before and during their exam periods. The blood samples were then assayed for cell count by using the routine method (complete blood count). Data were analyzed and compared with pre-examination results. There was no significant different in white blood cells, lymph counts and hemoglobin levels. On the contrary, a significant increase in platelet count was observed. The academic examinations in medical technology faculty are stressful enough to aggravate changes in platelet counts, even in healthy students.

**KEYWORDS:** white blood cell, lymph, hemoglobin, platelet, stress.

**INTRODUCTION**

Overcoming stress aids in allowing the immune system to become stable, although getting completely rid of stress is almost impossible. Due to the close interaction and communication between the immune system and the brain, it is not surprising that psychological upsets can consequent in physical symptoms, and sometimes even illnesses. Research has proven that exposure to chronic social conflict leads to high levels of stress, and, ultimately, disturbances to the immune system. It comes into view that chronic stress is capable of reducing the immune system's ability to defend the body from illnesses. [1]

Internationally, several studies have verified the effect of stress on different human body systems, including the immune system, though very few have been implemented in Arab countries. For that reason, the aim of this current study is to assess the influence of examination stress on the immune system, focusing on white blood cell counts. The reason why this study is centered on WBCs is because the rise of WBC counts is a useful and convenient signal to confine inflammatory responses. Another reason is that a complete blood count (CBC) test is inexpensive compared to other inflammatory markers like interleukin-6 (IL-6). Moreover, it is highly sensitive to C-reactive protein (CRP), and is regularly investigated during a routine health check up in various countries. [1, 2]

A study carried out in 2012 in the University of Al-Qadisiya, Faculty of Veterinary Medicine, established the effects of stress caused by academic examination on some physiological parameters. The investigation did not find significant differences on RBCs, PCV, WBCs, eosinophils and neutrophils among the studied groups. However the study did find notable effects on systolic blood pressure ($p \leq 0.05$) and also on students’ heart rates ($p \leq 0.05$) between groups. [3]

Most researches to date have intended to focus on the distribution of leukocytes and mast cells, rather than NK. One of these studies is an inquiry fulfilled to investigate the effects of academic examinations on total leukocytes count, neutrophil counts, lymphocytes and monocytes. The samples were drawn from 58 females. The study reported that examination stress caused significant increase in circulating blood neutrophil counts alongside slight decrease in circulating blood lymphocyte counts.

Seasonal variations affect the considerations of the results. Academic stress in the summer seemed to have a greater influence on the absolute lymphocyte counts than in winter. This was accompanied by a great increase in prolactin levels in summer compared with winter. On the
contrary, cortisol levels elevated more in winter than in the summer.[4]

Another study indicated that women are more likely to display negative immunological changes after marital stress than men. This was evidenced by their higher tiers of antibody to latent Epstein-Barr virus during the reactivation phase of the virus.[5]

Short-term studies, like those previously mentioned, do not show subtle change over time. A research carried out in a secondary school, Jammu, in India in 2015, compared its final results with pre-examination results. The blood samples that were taken during examinations showed a considerable decrease in eosinophil, basophil, lymphocyte and monocyte counts. Increase in platelet and neutrophil counts was also observed. No significant changes were seen in the counts of RBC.[6]

Different researchers have measured the effect of examination stress in a variety of ways. A study investigating the effect of academic stress on serum cortisol level and CD4 cell counts in young male students at Igbinedion University, in Okada, Nigeria, following results. Examination stress inhibits the proliferation of CD4 cells with the elevation of serum cortisol as a possible mediator.

It is largely recognized that chronic stress can lead to partial inhibition of the regulation of the immune functions.[9]

Generally, it has been accepted that T-lymphocytes, predominantly CD4 T-lymphocytes, play a central role in immune response. Cellular immune deficiencies lead to increased susceptibility to infection from viruses, fungi, intracellular bacteria and protozoa.[8]

Prolonged strenuous exercise has been acknowledged to decrease the number of T-lymphocytes and a more pronounced decrease in type I T-cells, which might be linked to high levels of plasma cortisol[1] as well as adrenaline.[9]

So far, much research has been descriptive in nature. An inquiry on the parameter of students’ blood cells in a Pakistani medical college explained whether exams in medical schools are stressful enough to produce such changes. A rise in systolic blood pressure by an average of 15mmHg was observed in 88% of the students in comparison to pre-examination readings. Significant changes during examination periods were also recorded in white blood cells. More considerable alterations were observed in basophil, eosinophil, lymphocyte and monocyte counts. Regarding RBCs, no significant changes were acknowledged and the hematocrit was also not disturbed.[9]

Physiological studies have shown that stress, from any source, is able to influence the endocrine, hematopoietic and immune systems. Cytokines and cortisol seem to have an important role in communicating between these systems.[12] Stress of academic examinations also noticeably affects the erythron variables. There is an increase in the number of large RBCs with increased haemoglobin, which cannot be explained by shifts of fluid out of the intravascular space, concentrating non-diffusible blood constituents.[10]

Additionally, it has been suggested that stress-induced pro-inflammatory cytokine production may stimulate the proliferation of hematopoietic cells.[11]

A recent study, published in 2017, examined the effect of pre-examination stress on autonomic, hematological, cognition endocrine and olfactory sensitivity in university students. The findings showed significant increase in cortisol levels observed in both males and females during the pre-examination period. Furthermore, neutrophil responses increased as well as the responses of monocytes, which elevated in males and females. On the contrary, lymphocyte and eosinophil responses decreased in both genders. Basophil responses significantly decreased in females, but did not show considerable change in males. No significant changes in olfactory sensitivity in both males and females.[12]

Recent work by Venkappa S Mantur and Dr. Vasudeva Murthy focusing on red blood cells was carried out in both absence and presence of examination stress. This study included 150 students, 76 of whom were male and 74 were female, accounting to 50.66% and 49.33% of the study population respectively. Of the 76 male students, 43 (56.57%) portrayed normal RBC count while 28 (36.84%) showed increased counts of RBCs in the absence of exam stress. Meanwhile, the analysis of RBC counts for the same group of male students under stress showed normal counts in 41 (53.94%) students and increased counts for the same group of male students under stress showed normal levels of RBC whereas the other 17 (22.97%) showed decreased levels. The main aim of this study is to evaluate whether examination stress can affect the white blood cell counts of students in the faculty of medical technology.

METHODS AND MATERIALS
40 healthy students (aged 18-23 years; average 20.5 years) volunteered to partake in our research from the faculty of medical technology at the University of Zawia. Students suffering from chronic diseases, such as asthma, etc. were disregard, as well as those on long-term medications and drugs like hormones, etc.

Sample Procedure
The first blood sample was withdrawn during the resting period, three months prior to the start of examinations.
whereas the second blood sample was taken just before the start of the students’ difficult exam, for example biochemistry, immunity and serology, etc. EDTA tubes were used for blood collection and sterile, non-toxic, pyrogen-free, disposable syringes were used to withdraw the blood. The blood was then analyzed by Mindray BS-200 chemistry analyzer, which is a discrete, random-access, fully-automated analyzer. The machine has 40 positions for samples, reagents (dilatants, rinses, lyses, probe cleanser and E-Z cleanser), control and calibrator, which are components of a system and feature automatic probe cleaning, liquid level detection and collision protection. It has a reversed optic system with eight wavelengths (340–670 nm). Values obtained during examinations were compared with those taken prior to look for changes of cell counts. A questionnaire was also prepared and given to students participating in the investigation. The survey contained both close- and open-end questions with simple words easily interpreted and answered by students in order to obtain symptoms and reasons of tension.

**Data Analysis**

In order to analyze changes in WBC count, lymph cells count, hemoglobin level and platelets count, SPSS software version 21 was carried out using (ANCOVA). All tests were two-sided, and $P$ values $>$ 0.05 were considered to indicate statistical significance.

**RESULTS**

The main aim of this study was to evaluate the affect of examination stress on the white blood cell counts of students, as well as to investigate the influence of examination stress of lymphocyte counts, hemoglobin contents, and platelet levels. 40 healthy, aged 18-23 years, were studied. Data was analyzed and the results from both study periods (before and during examination) were compared. Among the 40 students enrolled, there was a significant increase in platelet count ($p>0.05$).

### Table 1: WBC counts in the studied population.

<table>
<thead>
<tr>
<th>Period</th>
<th>Mean</th>
<th>N</th>
<th>Std. Deviation</th>
<th>% of Total Sum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rest</td>
<td>7.0000</td>
<td>20</td>
<td>1.66702</td>
<td>50.8%</td>
</tr>
<tr>
<td>During</td>
<td>6.7800</td>
<td>20</td>
<td>1.99278</td>
<td>49.2%</td>
</tr>
<tr>
<td>Total</td>
<td>6.8900</td>
<td>40</td>
<td>1.8168</td>
<td>100.0%</td>
</tr>
</tbody>
</table>

![WBC Count](image)

**Figure 1:** WBC Counts in the Studied Population, the mean ± SD of WBC. In the graph, blue indicates the mean and red indicates the standard deviation. No significant difference between the two periods was seen.

### Table 2: Lymph counts among the study population.

<table>
<thead>
<tr>
<th>Group</th>
<th>Mean</th>
<th>N</th>
<th>Std. Deviation</th>
<th>% of Total Sum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rest</td>
<td>2.1700</td>
<td>20</td>
<td>0.47914</td>
<td>50.9%</td>
</tr>
<tr>
<td>During</td>
<td>2.0950</td>
<td>20</td>
<td>0.49681</td>
<td>49.1%</td>
</tr>
<tr>
<td>Total</td>
<td>2.1325</td>
<td>40</td>
<td>0.48325</td>
<td>100.0%</td>
</tr>
</tbody>
</table>
Figure 2: Lymph Counts Among the Study Population, the mean ± SD of lymphatic cell counts. In the figure, red indicates S.D, while blue indicates the mean. No differences were interpreted among the different periods.

Table 3: Haemoglobin levels among the study population.

<table>
<thead>
<tr>
<th>Period</th>
<th>Mean</th>
<th>N</th>
<th>S.D</th>
<th>% of Total Sum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rest</td>
<td>10.8800</td>
<td>20</td>
<td>1.17141</td>
<td>49.4%</td>
</tr>
<tr>
<td>During</td>
<td>11.1350</td>
<td>20</td>
<td>1.46045</td>
<td>50.6%</td>
</tr>
<tr>
<td>Total</td>
<td>11.0075</td>
<td>40</td>
<td>1.31313</td>
<td>100.0%</td>
</tr>
</tbody>
</table>

Figure 3: Haemoglobin Levels among the Study Population. the mean ± SD of haemoglobin levels. In the figure, blue indicates mean and red indicates standard deviation. There was no significant change amid the two periods.

Table 4: PLT count in the study population.

<table>
<thead>
<tr>
<th>Group</th>
<th>Mean</th>
<th>N</th>
<th>S.D</th>
<th>% of Total Sum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rest</td>
<td>233.9000</td>
<td>20</td>
<td>60.00693</td>
<td>48.6%</td>
</tr>
<tr>
<td>During</td>
<td>247.8500</td>
<td>20</td>
<td>55.10972</td>
<td>51.4%</td>
</tr>
<tr>
<td>Total</td>
<td>240.8750</td>
<td>40</td>
<td>57.30404</td>
<td>100.0%</td>
</tr>
</tbody>
</table>
DISCUSSION
This study found that individuals undergoing the normal stress of academic examinations exhibited indistinct differences in WBC count, lymph cells count, platelet count and hemoglobin level. Prior studies, which noted the importance of examination stress on the immune system, showed that academic stress in the summer seemed to have a greater influence on the absolute lymphocyte counts than in winter.[4] As mentioned in the literature review, a study in Jammu, in India in 2015, compared its final results with pre-examination results.[6]

The blood samples that were taken during examinations showed a considerable decrease in eosinophil, basophil, lymphocyte and monocyte counts. Previous studies evaluated significant changes in white blood cells. More considerable alterations were observed in basophil, eosinophil, lymphocyte and monocyte counts.[13] This current study indicated that WBC appeared to be unaffected by stress. This result is similar to a previous study in the University of Al-Qadisiya, which demonstrated that there were no significant changes on WBC count.[3] However, the findings of the current study do not support the previous research at a Pakistani medical college, which recorded significant changes in white blood cells during examination periods.[9]

The outcomes of the results in this study regarding lymph showed no significant change contrarily to that of another study, which found a reduction in lymphocyte and monocyte counts.[13] One unanticipated finding was the platelet counts. This outstanding change in the present study also suggests that the emotional state of the student must be considered when interpreting platelet-aggregation. Although the link is not proven, strong epidemiologic data suggests that stress may be an etiological factor in the development of coronary artery disease and myocardial infarction.[14, 15] Catecholamines, normally secreted in abundance during stress, are known to be potent platelet-aggregating agents in vitro.[16, 17] Physiological changes in platelet function serve to inhibit platelet reactivity. This type of reaction might be of protective value because during stress, the heightened secretion of catecholamines might lead to inappropriate intravascular platelet aggregation, if the platelets were sensitive.

This study did not detect any evidence for change in the hemoglobin content, in contrast to earlier findings which stated that the stress of academic examinations noticeably affects the erythrocyte counts. There was an increase in the number of large RBCs with increased haemoglobin levels, but not enough to cause changes in white blood cell, lymph and hemoglobin levels.[10]

REFERENCES
student pilots. Aviation Space Environmental Medicine. 64: 116 – 120.