# The Changes in Some Hematological Parameters among University Students Due to Stressful Conditions During and after Examinations Period

**Jehad F. Alhmoud[1], Husni S. Farah[2], Talal Al-Qaisi[1], Wala'a Hamdan[3], Ghaleb A. Oriquat[4], Ali Atoom[1], Khalid M. Al-Qaisi[1]**

*[1]Assistant professor. [2]Associate professor, [3]Tutor .[4]Professor, Department of Medical Laboratory Sciences, Faculty of Allied Medical Sciences, Al-Ahliyya Amman University, Zip code (19328) / Amman, Jordan.*

## Abstract

**Background:** Several studies demonstrated that stress can cause an obvious change in the hematological parameters in healthy individuals. In the current study, we determined whether the stress during the exams can produce some hematological changes and how long this effect may take after the end of the exam period.

**Methods:** Seventy (n= 70) male and sixty (n=60) female student of similar height, weight and age were selected from the department of medical laboratory sciences, Al-Ahliyya Amman university. All students were first had a preliminary medical checkup. Students excluded from the study include those who had a chronic disease and any health issues or habits affecting the studied parameters, such as smoking; fever and high blood pressure. All study participants were between the age group of 20-22 years. Blood samples were taken three weeks before the exam, during and 72 hours after the end of the exam period. Estimations of red blood cells (RBC), white blood cells (WBC), hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), neutrophils, lymphocytes and platelets were accomplished.

**Results**: Compared with pre-examination results the blood samples taken during exams showed a significant decrease in the readings of lymphocytes ($p < 0.01$) , MCV and MCH ($p < 0.05$). Significant increase in Neutrophil ($p < 0.05$) and platelets ($p < 0.05$). Readings taken after 72 hours of the end of exam period showed significant decrease in white blood cell count ($p < 0.01$) and further significant increase in the platelet count ($p < 0.001$). No significant changes were observed in the readings of Hemoglobin, Red Blood Cell count (RBC), hematocrit and mean corpuscular hemoglobin concentration ($p > 0.2$).

**Conclusion:** This study concluded that the stress due to examinations is enough to alter certain hematological parameters. The effect of stress on platelets and WBC remained until the end of the exam period. As the increased platelet count may cause health problems for a person, from bleeding problems to the formation of various clots. We do not know for sure how long the increase in platelet and decrease in white blood cells continue as this may cause health problems for people subject to permanent stress.

***Kew words**: Examination stress, hematological parameters, university students.*

**Corresponding author:**
**Dr. Husni Farah**
Associate professor**,** Department of Medical Laboratory Sciences, Faculty of Allied Medical Sciences, Al-Ahliyya Amman University, Zip code (19328) / Amman, Jordan;
Email: h.farah@ammanu.edu.jo
Mobile No.: 00962796535979

## Introduction

In any educational system, improving examination performances of students and reducing their failures is the main goal. In order to achieve better outcomes in any educational system, it is necessary to investigate the factors that contribute to the student's exam performance. Stress is one of the main notable factors that produced by heavy academic load, stress of everyday exams and

absence of relaxation [1]. Physiological studies have recommended that stress of any kind will have a significant impact on hematogenic and endocrine systems[2]. Stress can also change the blood cell parameters include; neutrophils, platelets, eosinophils, monocytes basophils and lymphocytes in healthy individuals as demonstrated in physiological studies [3]. Moreover, several studies prove that the immune system is affected by both stress and emotional reactions [4,5]. However, complete blood counts, including the hematopoietic system, leukocyte profile or biochemical markers of stress are all mostly used to recognize possible changes in the immunological system [6]. Some other published study demonstrated that stress can impact the number and quality of the cells, such as; leukocyte, platelet and erythrocyte populations which need constant replication to maintain their counts, as in the normal situation these cells characterized a constant turnover of cells [7,8]. Stress due to university examinations significantly increases the readings of red blood cells and hemoglobin [3].

A published study on the occupational noise that cause a type of stress on the individuals exposed to noise for several years. Exposure to noise can lead to increase stress, tissue dysfunction, and cause changes in the normal process of secretion of the body's hormones, thus resulting in significant effects on blood parameters and health[9]. This study showed that some hematological parameters such as; MCV, MCH, FBS, and MCHC were declined. While other parameters including cholesterol, RBC, WBC, Hb, and Hct were induced compared with the administrative workers. Therefore, it is obvious that there are noteworthy changes on blood parameters of these workers, which can have harmful effects on their health in the future [10]. The literature demonstrated a study mainly focused on observing the changes in hematological parameters and serum cortisol levels after examinations conducted in the medical profession on 1st year medical students. This result, suggesting that stress is responsible for the changes of these parameters. This published study recommended that proper care and support from both the faculty and the parents is provided, as it is essential for the improvement of the immune system and allow the students to adapt sufficiently to exam stress [11]. However, another study focused on investigating the effects of noise and chemical stress on hematological and biochemical parameters of the human body through comparing two groups of people involved in maintenance and transport related work and the other group involved in more responsible duty, working under a continuous chemical and noise stress environment. The results clarify a clear influence of chemical and noise stress on the human body in terms of hematology and biochemistry through showing an increase in total serum protein level, serum urea level, and platelet count in stress exposed group beside a significant decrease in serum triglyceride level in the same group. In addition, it was observed that there is statistically insignificant increase in the count of blood Red blood cells (RBC), White blood cells (WBC), Hematocrit, Mean corpuscular hemoglobin concentration (MCHC) and mean cell volume (MCV) in stress exposed group than normal group [12]. As this project helps to understand the role of important hematological parameters in monitoring the stressful conditions, which could affect the student behavior during the exam. Better understanding of the quality of life issues could assist the students to manage their stress in order to perform better in their exams. Our study involved one hundred thirty students n= 130 students from both sexes. The blood sampling was conducted before, during and after the exam. However, the study of hematological is essential to assess the physiological status in human. For a fully understand the changes in the mentioned parameters, we have to know the baseline physiological parameters in order to compare the results with the students under stress.

Generally, we are expecting that the parameters investigated during the exam will be different from the parameters measured before or after the exam. The current study focused on the undergraduate students in the 3rd year and our main target is to observe the changes in hematological parameter including Red blood cells (RBC) count, haematocrit (Hct), hemoglobin (Hb), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), white blood cells (WBC) count and platelets.

## Methodology

### Study design and subjects

The present study was conducted at Department of Medical Laboratory Sciences, Faculty of Allied Medical Sciences, at Al-Ahliyya Amman University/ Amman. A random group was chosen from the third

year students in the College of Medical Sciences. All students were first had a preliminary medical checkup. Students excluded from the study include those who had a chronic disease or any health issues or habits affecting the studied parameters, such as smoking; fever and high blood pressure. Finally, seventy (n= 70) male and sixty (n=60) female students of similar height and weight were included in this study between the age group of 20-22 years. All participated students were informed about all the procedures and outcomes of the study.

**Complete blood profiles**

Five milliliters of blood were taken from each student. It was transferred into lavender vacutainer, containing Ethylenediaminetetraacetic acid (EDTA) as an anticoagulant. Blood count was performed on an automated hematology analyzer (Sysmex XP-300) at Sysmex Corporation, Japan. Each count presented the data of white blood cell count (WBC), red blood count (RBC) platelets count (Plt), mean corpuscular volume (MCV), measurement of hemoglobin (Hgb), hematocrit (Hct), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC). The determination of blood count was performed for each student three weeks before exam period, during exams and 72 hours after the end of the exam period.

**Statistical Analysis**

The data were analyzed and determine the relationship between various parameters, The un-paired t-test was applied to determine statistical significance. Results were presented as means ± SD. Values <0.05 were regarded as statistically significant.

**Results and Discussion**

This study was carried out to investigate the changes in hematological parameters including red blood corpuscles (RBCs), white blood cells (WBCs), haematocrit, haemoglobin and related RBC indices. A total 130 undergraduate students in the third year medical laboratory sciences were

**Table 1**. Changes in White blood cell count, Relative (%) lymphocyte count, relative (%) neutrophil count and platelets count were determined before the exam, during exam and 72 hrs. after the end of exam period. Results show the mean value of 130 students. Significant difference between the count before, during and 72 hrs. after exam is indicated as not significant (NS), *p< 0.05, ** p<0.01 and ***, p < 0.001.

| Hematological parameter | Before exam ±SD | During exam ±SD | 72 hrs. after exam end of period ±SD |
|---|---|---|---|
| White blood cells × 103 / µl | 8.5 ±2.5 | 8.8± 3.2 | 5.5 ± 1.9 |
| Relative lymphocytes count (%) | 33.6 ±4.2 | 26.8 ± 2.2 | 31.7 ±3.6 |
| Relative neutrophil (%) | 61.3 ±3.8 | 68.2± 4.7 | 63.3 ±2.7 |
| Platelets count × 106 µl | 275.6 ±42.4 | 322.1± 35.5 | 365.8±48.2 |

participated in this study. All study participants were between the age group of 20-22 years. The results from this study revealed a significant decrease in WBS count (5.5 × 103 /µl ± 1.9, p < 0.01) 72 hrs. after the end of exam period exam compared with that before exam period (8.6 × 103 /µl ± 2.5) as shown in (Table 1). This such decrease in WBC agreed with the studies of Bopda B et al[13], Qureshi F et al [14]and Johansson G. et al [15] and contradicts with the reports of Roohi N, Irfan S[16] and Venkappa S, Vasudeva M.[17] The lymphocyte counts

(%) showed significant decrease during the exam period (26.8 ±2.2, p <0.01) whereas the neutrophil counts (%) increased significantly during the exam period (68.2 ± 4.7, p<0.05) compared to that before the exam which agreed with the reports of Viktoriya M. et al [18] and Nakamura H. et al[19]. The decrease in the absolute numbers of lymphocytes cells has been associated with chronic stress which results in redistribution of lymphocyte cells and caused them to adhere to the endothelium (Nakamura H. et al )[19] . Suleiman D. et al [20] found an increased number in neutrophils in case of chronis stress and attributed this to the increase in both acute phase reactants and the functions of TNF-alpha. It is very interesting to find a significant increase (322.1× 106 /µl ±35.5, P<0.05) in platelet count (Table 1) during the exam and there has been a further high significant increase (365.8× 106 /µl ± 48.2, p<0.001) in their count 72 hours after the end of the exam period. Their counts returned to what they were before the exam (Table 1) as none of the previous studies ( (Bhaskara K.[3] and Johansson G. et al.[15]) recorded this result, but all were satisfied with investigating the effect of stress before and during the exam period. We believe that one of the reasons for the persistence of high platelets after the end of the exam period is the continued rise in the adrenaline hormone, which increases as a result of stress. Lande K. et al [21] showed a close relationship between the hormone of adrenaline and the increase in platelet count. We think that students during exam may suffer from sleep deprivation and there is some evidence (Bangasser D. et al [22] and Sharma N, Gupta V [23]) that sleep deprivation and the increased secretion of cortisol, which is linked to the extent of stress, both suppress the immune system, which in turn explains a decrease in efficiency of the immune function causing a decrease in both WBC and lymphocyte counts. Also WBC and Lymphocytes possess receptors for adrenaline as its plasma levels increase in response to stress. Adrenaline reduces the flow of white blood cells, causing a redistribution of WBC and lymphocytes between the blood and other immune compartments ( Dhabhar F et al. [24]) causing the free circulating WBC and lymphocytes to decrease. The present study showed a significant increase in neutrophils during exam period that consistent with the results of Bopda B. et al [13]and Qureshi F. et al[14] . We think that the type of lifestyle, dietary system and family conditions may explain the variable change in the level of stress in those students, which is reflected in the results of hematological parameters. Pre-examination stress could be a quite common condition among University students. Several studies were carried out and the changes that occur as a result of stress have not been identified fully.

**Table 2.** Change in Hemoglobin, Red blood cell count, Mean corpuscular volume MCV, Hematocrit percentage, Mean corpuscular hemoglobin MCH and Mean corpuscular hemoglobin concentration MCHC were determined before the exam, during exam and 72 hrs. after the end of exam period. Results show the mean value of 130 students. The significant difference between the count before, during and 72 hrs. after exam is indicated as not significant (NS), *p< 0.05.

| Hematological parameter | Before exam ±SD | During exam ±SD | 72 hrs. after exam end of period ±SD |
|---|---|---|---|
| Hemoglobin (g/dl) | 14.4 ±2.2 | 14.7 ±2.3 | 14.4 ±1.9 |
| Total red blood cell count × 106 µl | 5.1 ±3.2 | 5.6 ±3.4 | 5.2 ±2.8 |
| Mean corpuscular hemoglobin MCH (pg) | 27.3 ±2.2 | 24.4± 2.3 | 27.7 ±2.5 |
| Hematocrit (%) | 43.3 ±3.7 | 43.7 ±3.2 | 43.1 ±2.9 |
| Mean corpuscular volume MCV (fL) | 82.5 ±2.4 | 74.3±3.8 | 77.5 ±5.3 |
| Mean corpuscular hemoglobin concentration MCHC (g/dl) | 35.1 ±0.81 | 35.3 ±0.98 | 35.4 ±1.1 |

The present study demonstrated a significant decrease in the level MCH and MCV during the exam period (24.4 ± 2.3, p<0.05, 74.3 ± 3.8. P< 0.05) respectively which is consistent with the report of Eun et al [12], this such decrease was explained by Hisham W and Mohammed A[25] that deficiency in Glutathione reductase which plays an important role in protecting biological cell membranes of RBC against oxidative damage that results from stressful conditions. Their counts returned to what they were before the exam period (Table 2), which undoubtedly confirm the effect of stress on these parameters. There was no change in the levels of Hb, MCHC and hematocrit all through the study (Table 2) which contradicts the studies carried out by Bopda B et al [13]and Qureshi F. et al [14] that recorded a decrease in the levels of these parameters during the examination session.

In this study it was observed that stress affects significantly more female students (75%) than a male one (53.3%). This corresponds to previous report of Lande K. et al [21] on the impact of stress on women more than men as the emotional nature of a woman plays a role in a woman's sense of stress and anxiety more than men.

## Conclusion

A large number of students of the university were affected by examination Stress especially during the exam. The study sample participated in this study showed stress that affects some of the hematological parameters. Our study investigated the effect of stress after the end of the exam period, which none of the previous studies addressed that. This study showed that platelets increased and WBC decreased after the end of the exam period and we do not how long they can take to return to normal level. This result about platelets may explain the fact that the stressful people as exposing themselves to continuous rise in platelets and the risk of high blood clot.

**Author Contributions**: Jehad .Al-Homoud and Husni Farah conceptualized and supervised the article. Jehad .Al-Homoud wrote the initial draft and revised subsequent drafts. Talal .Al-Qaisi ,Ali Atoom , ghaleb Oriquat and Khalid Al-Qaisi analyzed, reviewed and edited the manuscript prior to submission. Wala,a Hamdan did the practical work in the lab. All authors have read and agreed to the published version of the article.

**Funding:** This research received no external funding.

**Ethical statement**: Based on the Ethics Committee's decision of Al Ahliya Amman University/Jordan dated 08/10/2019, reference number 1/1/2019/2020 (578) it was agreed to carry out the experiments necessary to complete this research

**Conflicts of Interest:** The authors declare no conflict of interest.

## References

1) Hettiarachchi M, Fonseka C, Gunasekara, P. Jayasinghe, D. Maduranga D. How does the quality of life and the underlying biochemical indicators correlate with the performance in academic examinations in a group of medical students of Sri Lanka? Med Educ Online, 2014;19 (1): 7- 9

2) Dorshkind K , Horseman N. Anterior pituitary hormones, stress, and immune system homeostasis. BioEssays. 2001,

3) Bhaskara K. Assessment of physiological parameters in medical graduates under examination stress. .J App Med Sci. 2017; 5(6A):2074

4) Christopher P F, Baldwin W. Early-Life Stress and Adult Inflammation. Current Directions in Psychological Science. 2014; 23(4) : 277-283

5) Morey J, Boggero I, Scott A, Segerstrom A. Current directions in stress and human immune function. Curr Opin Psychol. 2015; 1 (5): 13–17.

6) Leandro C. Effect of L-Glutamine on the number of blood leukocytes and on the phagocytic function of macrophages of stressed rats. Rev. Nutr 2006; 19(4): 437 - 444

7) Deepak K, Padmapriya K , Srinivasa M. Erythrocytes and Platelets: A Critical Analysis of their Ontogenic Relationship through Automated Parameters. J Clin Diagn Res. 2017; 11(5): EC05–BC08.

8) Valdenilson R R, Hugo André L M, Marcelo T V, Simone N F, Severino Marcos OC; Bruno HA et al. Alice Andrade. Hematological and immunological effects of stress of air traffic controllers in northeastern Brazil. Revista Brasileira de Hematologia e Hemoterapia. 2011; 33 (3): 195 -

201.

9) Marita KG, Kandregula S. Acute noise exposure effect on certain hematological parameters in tailors: A pilot study. Int J Biol Med Res. 2012 ;3(2): 1529-1531

10) Mohammadi H. Alimohammadi I, Roshani S, Pakzad R, Abdollahi MB, Farhang SD. The Effect of Occupational Noise Exposure on Blood and Biochemical Parameters: A Case Study of an Insulator Manufacturer in Iran. Electron. physician. 2016; 8(1):1740-1746

11) Ali I , Yasmeen N . Corelation of stress with hematological parameters and serum cortisol levels before and during examination in 1 st MBBS Students. Int. J. Physiol. 2018; 6 (4): 111- 115.

12) Eun HN, Suyoung IK, Seon IC, Han IC. Complete Blood Count Reference Intervals and Patterns of Changes Across Pediatric. Ann Lab Med. 2018; 38(6): 503-511

13) Bopda B O, Sumbele O, Nji L. The Effect of Stress on Cardiovascular Parameters among University Students in Buea during and Out of Examination Periods. J. Adv. Med. Med. Res. 2019, 28(12)1-4.

14) Qureshi F, Alam J, Khan M, Sheraz G. Effect of examination stress on blood cell parameters of students in a Pakistani medical college. J. Ayub Med. Coll. Abbottabad. 20011; 4(1):20-22.

15) Johansson G, Laakso M, Peder M, Karonen S. Examination stress decreases plasma level of luteinizing hormone in male students. Psychosom. Med. 1988;50(3):286- 294.

16) Roohi N, Irfan S. Haematological and hormonal response of traffic policemen. Pak J Sci. 2009; 61:20–5.

17) Venkappa S, Vasudeva M. Effects of academic stress on leukocyte subset distribution in humans. J. Chinese Clin. Med. 2010; 5(9): 41 - 19.

18) Viktoriya M, Maren C, Nicole D, Melanie E, Jürgen D, Stefan D, Oliver T, Thomas K, Carsten W. Impact of chronic and acute academic stress on lymphocyte subsets and monocyte function. PLoS One. 2017;16(11):1 - 19

19) Nakamura H, Nagase H, Yoshida M, Ogino K. Natural killer (NK) cell activity and NK cell subsets in workers with a tendency of burnout. J Psychosom Res. 1999;46(6):569–78.

20) Suleiman D, Abdullah A, Mahmut B, Aslıhan O, Mehmet G, Mehmet C, Özlem D, Aytekin S. Neutrophil-lymphocyte ratio in patients with major depressive disorder undergoing no pharmacological therapy. Neuropsychiatr Dis Treat. 2015; 27(11) :2253-8.

21) Lande K, Gjesdal K,Fønstelien E, Kjeldsen S, Eide I. Effects of adrenaline infusion on platelet number, volume and release reaction. Thromb. Haemost. 1985; 54(2):450-453.

22) Bangasser D A, Eck S, Telenson A, Salvatore M. Sex differences in stress regulation of arousal and cognition. Physiol. Behav.2018; 1(187):42-50.

23) Sharma N, Gupta V. Effect of examination stress on hematological and hemodynamic," Interational J. Pharmacol. Physiol.2015;15(2):153-156

24) Dhabhar F, Miller A H, McEwen B S, Spencer R L. "Effects of stress on immune cell distribution. Dynamics and hormonal mechanisms. J. Immunol. 1995; 15(154):5511-27.

25) Hisham W, Mohammed A. The effect of oxidative stress on human red cells glutathione peroxidase, glutathione reductase level, and prevalence of anemia among diabetics. N Am J Med Sci. 2011;3(7): 344–347.