Journal of Medical and Health Studies
ISSN: 2710-1452
DOI: 10.32996/jmhs
Journal Homepage: www.al-kindipublisher.com/index.php/jmhs

JMHS
AL-KINDI CENTER FOR RESEARCH AND DEVELOPMENT

| RESEARCH ARTICLE

The Effect of Psychological Stress on the Level of Hematological Parameters and Cortisol Hormone among Students in the University

Muhammad Riyadh Alkenany[1] ✉ Rusul Ali Al-Masaoodi[2], Ahmed A. AL-Safar[3] and Noor Ali Abdul Imams[4]
[1]Department of Medical Physics, Faculty of Medical Applied Sciences, University of Kerbala, Karbala, 56001, Iraq
[23]Department of Environmental Health, Faculty of Medical Applied Sciences, University of Kerbala, Karbala, 56001, Iraq
[4]College of Veterinary Medicine, University of Karbala, Karbala, 56001, Iraq
Corresponding Author: Muhammad Riyadh Alkenany, **E-mail**: mu7066941@gmail.com

| ABSTRACT
Stress is a mechanism used by individuals to deal with problems and challenges that might arise from the inside or outside. The brain and the architecture of the body are directly impacted by stress. Among other things, psychological issues, health issues, sadness, anxiety, discomfort, and exhaustion can result from an inability to adjust to a stressful circumstance. This research was conducted on students of University to find out the effect of psychological stress on some immunological and hematological parameters. The research was conducted on twenty specific college students and blood samples were drawn from them in three stages. The first stage is the stage before exposure to psychological stress and during the rest period. The second stage is drawing blood from students while they are exposed to psychological stress (during exam times). The third stage is drawing blood from students after the psychological stress factor has passed and during the period of complete rest. The results showed a significant decrease in the value of the total white cell count and lymphocyte percentage and a significant increase in the percentage of neutrophil during the period of exposure to the factor of psychological stress as compared to the period before exposure to stress and then re-rise after the vanishing of stress-causing factor. On the other hand, the results showed a significant increase in the number of red blood cells and hemoglobin concentration during the period of exposure to psychological stress, in comparison with the two periods before and after the stress factor (at resting period) was removed. the results of the hormonal analysis showed a significant and clear increase in the concentration of cortisone during the period of exposure to psychological factor and as compared with the rest period before and after exposure to the psychological stress factor. For the first time in Karbala province, it has been proved that the research results showed clear influence of psychological factors on the immune status related to human health.

| KEYWORDS

Stress, University students, Cortisol, WBC, RBC.

| ARTICLE INFORMATION

ACCEPTED: 02 August 2025 **PUBLISHED:** 21 September 2025 **DOI:** 10.32996/jmhs.2025.6.4.11

1. Introduction

By causing physiological reactions to different stimuli, psychological stress upsets the organism's equilibrium. Certain stress-related hormones are produced and released as a result of this disruption, which also stimulates two important neuroendocrine pathways: the sympathetic-adrenal-medullary (SAM) system and the hypothalamic-pituitary-adrenal (HPA) axis (Ouda, S., *et al.* 2016). Emotional stress induces psychological reactions that alter multiple components of the neuroimmune-endocrine network, including the immune system, autonomic nervous system (ANS), pituitary and hypothalamus hormone levels, neuropeptides, and cytokines (Dhama, Kuldeep, *et al.*, 2019). Psychological stress resulting from stress due to studying in the educational system is one of the most common things among students, which greatly affects their academic level. The primary purpose of every educational system is to improve students' examination performance and reduce their failure rates. To improve results in any educational institution, it is vital to analyze elements that influence the student's exam performance. tension is one of the most

noteworthy variables generated by severe academic load, tension of everyday tests, and lack of leisure (Hettiarachchi *et al.*, 2014).

Stress is a widespread concern in people's lives. As mentioned by Kupriyanov and Zhdanov (2014), stress is presently seen as a necessary component of contemporary existence. To put it another way, stress is now an inevitable part of life, whether it be at work, school, home, or somewhere else. The term "academic stress" describes tension that develops in a learning environment. Stress related to school might endanger one's physical and emotional well-being. High levels of stress can lead to problems for anybody, including students, claim Lin and Huang (2014).

There are two types of stress related to academics: biological and psychosocial (cognitive, emotional, and social). The term "biological stress" refers to the physical reactions that arise from stress, such as headaches, elevated heart rate, and trembling legs, which are caused by the sympathetic nervous system and endocrine system. Second, psychosocial cognitive, or the person's cognitive response to stress, includes things like trouble concentrating, forgetfulness, emotions of worthlessness, bewilderment, desperation, negative thinking, decreased accomplishment, a sense of not enjoying life, and trouble making decisions. (Sarafino and Smith, 2014).

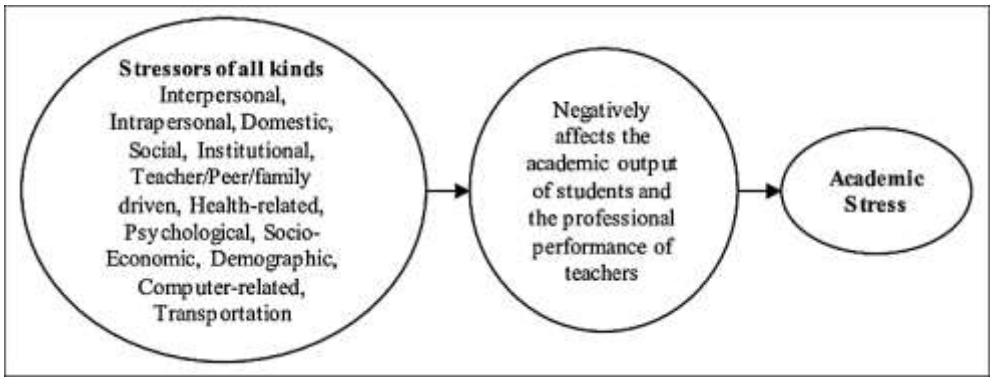

Figure 1: Conceptual framework for academic stress (Adom *et al.,* 2020)

Effective stress management is essential for immune system stability, even if it is nearly impossible to eradicate stress. Given the intimate and reciprocal relationship between the brain and the immune system, it is possible for psychological issues to cause physical symptoms and, in certain situations, contribute to the onset of somatic or immunological disorders. According to research, long-term exposure to social conflict is substantially linked to higher stress levels, which over time may cause immunological dysregulation. As a result, ongoing psychological stress may hinder the immune system's capacity to effectively defend against pathogenic threats (Elenkor and Chrousos, 2002). The complementary vitamin C and zinc could maintain a normal immune system function (AL-azawi and ALkenany, 2017).

The endocrine system releases hormones responsible for stress like adrenaline and cortisol when a situation is recognized as possibly harmful. Stress begins with the production of an alert, which the body recognizes and then releases the hormone adrenaline to increase the body's response to any unforeseen event. This leads the sympathetic and parasympathetic nervous systems to surge. The sympathetic nervous system begins to secrete noradrenaline or epinephrine (NE) at about ten times the normal rate, resulting in more epinephrine (E) than norepinephrine (NE). Hormone production is triggered by stress or panic, which prompts the hypothalamus to receive information from the brain. Among these hormones is cortisol, an anti-inflammatory that helps the body distribute its energy sources and can be interpreted as a stress indicator (Hellhammer *et al.*, 2009; Martinez *et al.,* 2009).

While cortisol is assumed to have an impact on the metabolic system, epinephrine is linked to depression, hypertension, and attention deficit hyperactivity disorder (ADHD). According to Sandman *et al.* (2011), stress hormones influence metabolic processes, including digestion, reproduction, development, and immunity. They also increase heart rate, blood pressure, and breathing rate. They also release stored energy into the muscles.

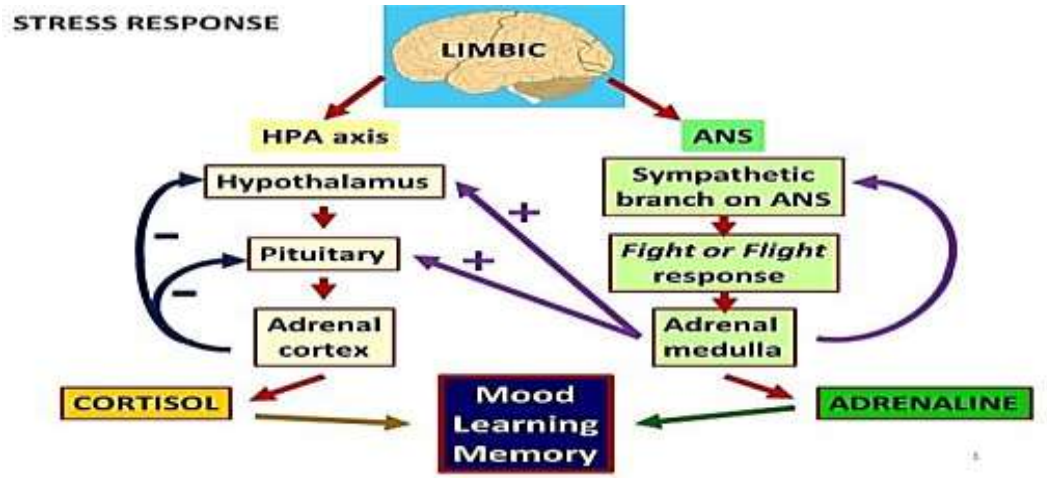

Figure 2: Diagram of the two major stress system (Banic and Compton, 2018)

2. Materials and Methods

The experiment was carried out at the university. Twenty healthy adult students in the same college were used in this study, their ages were ranged between 22-25 years old, and their weight was around 55-110 kilogram. blood samples were obtained from same student at three stages as follow: show figure 3.

Three stapes were done in this section, Stage one (pre psychological stress) of blood collection(PS) was in the rest period when they were absolutely resting without any psychological stress cause, then the Stage two (during psychological stress) of blood collection (DS) was during the psychological stress activator (exam). Finally, Stage three (post psychological stress) of blood collection (POS) was post the stress activator (exam) when the patient return to the resting period when they were absolutely at rest. The duration between the stages of blood collection was ranged between 7-10 days between each of the three stages of collection (Meryer et al., 1982).

2.1 Sample Collection

Blood samples were taken from the pupils once the experiment was over. Each subject had their venous blood extracted using disposable syringes. To avoid coagulation, the obtained samples were moved into EDTA and Gel tubes. After centrifuging the serum for 10 minutes at 3000 revolutions per minute (rpm), the serum samples were kept at -18°C until they could be examined further.

2.2 Hemoglobin and Immunological Estimation

In the hematology laboratory, EDTA blood was used to test the hematological parameters utilizing Mythic 18 (RINGELISA N CO.,Turkey). With EDTA anticoagulated blood, this fully automated hematology analyzer assessed the complete blood count (CBC).

2.3 Serum cortisol Assay

The Fincare™ fast test for cortisol measurement uses a comparative immunodetection mechanism and fluorescence immunoassay (FIA) technology. Fluorescently labeled anti-cortisol antibodies react with the cortisol molecules in the blood sample when the sample is placed into the test cartridge's designated well, forming antibody–antigen immune complexes.

2.4 Statistical Analysis

The Statistical Analysis System (SAS) software, version 9, was used to statistically analyze the experimental data. To assess significant differences between group means, a one-way analysis of variance (ANOVA) and the Least Significant Difference (LSD) test were used. P-values below 0.05 were regarded as statistically significant.

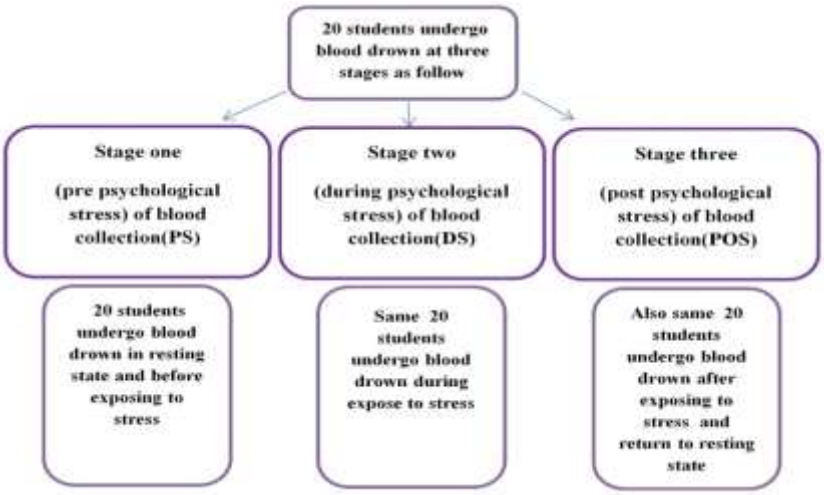

Figure 3: The design of the study

3. Results

Table 1 represent the effect of psychological stress on total leukocyte count (cellx103/mm3) and Lymphocyte (%) results of leukocytes and lymphocytes counts showed that the WBC count significantly decrease during stress and return to rise after diminishing of stress cause. From other hand, the lymphocyte percentage also decrease significantly during exposing to psychological stress and return to enhanced and elevate after disappearing of stress, but neutrophil (%) increase substantially during stress and return to decrease after diminishing of stress cause as show figures 4, 5 and 6.

Table 1: The effect of psychological stress on leukocyte count(cellx103/mm3) and Lymphocyte (%) in male and female students at university

Parameter	Pre stress	During stress	Post stress	LSD
Leukocyte count (cellx10^3/mm^3)	**9.49**±0.26 A	**7.61**±0.35 C	**8.74**±0.37 B	**1.01**
Lymphocyte (%)	**35.84**±0.251 A	**23.96**±0.296 B	**36.57**±0.605 C	**1.26**
Neutrophil (%)	**63.67**±0.911 B	**67.93**±0.957 A	**53.57**±1.275 C	**3.24**

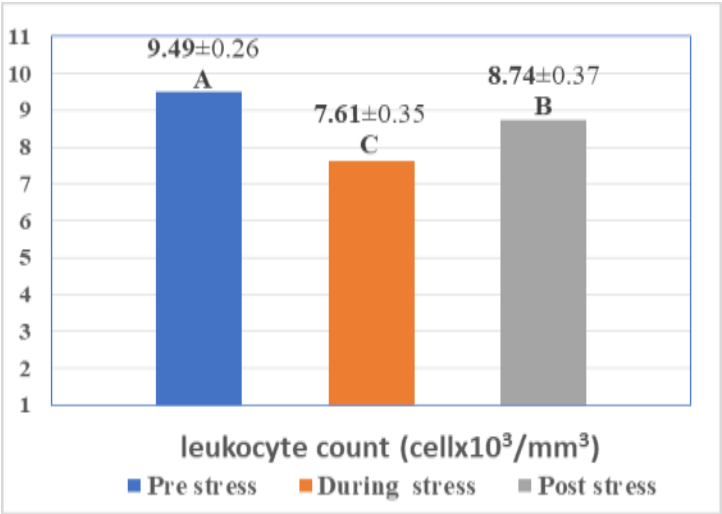

Figure 4: Mental Stress's Effect on Leukocyte Count (cells ×10^3/mm^3):

The statistically significant differences in leukocyte counts across the three research groups are depicted in this figure. The pre-stress and during-stress phases (A) showed a substantial rise, whereas the stress and post-stress phases (C) showed a significant difference. Nevertheless, there was no statistically significant change between the pre-stress and post-stress periods (B).

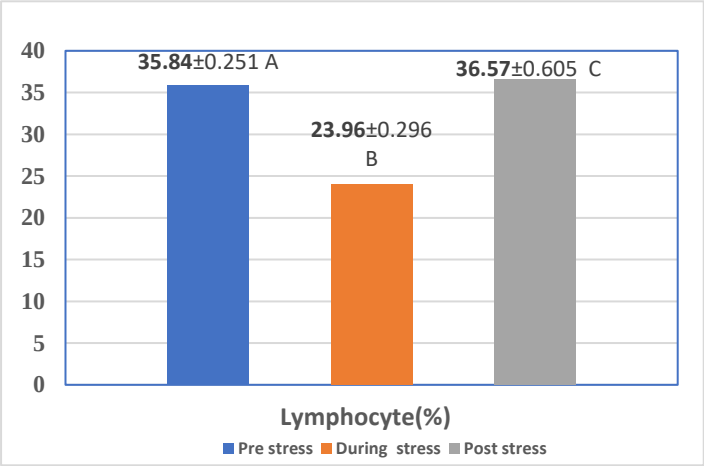

Figure 5: The effect of psychological stress this figure shows the between the Lymphocyte(%). this figure shows the significant difference between the Lymphocyte count in three groups of study(A)significant difference between pre stress and during stress , (B) non- significant difference between pre-stress and post stress, (C) significant difference between during stress and post stress.

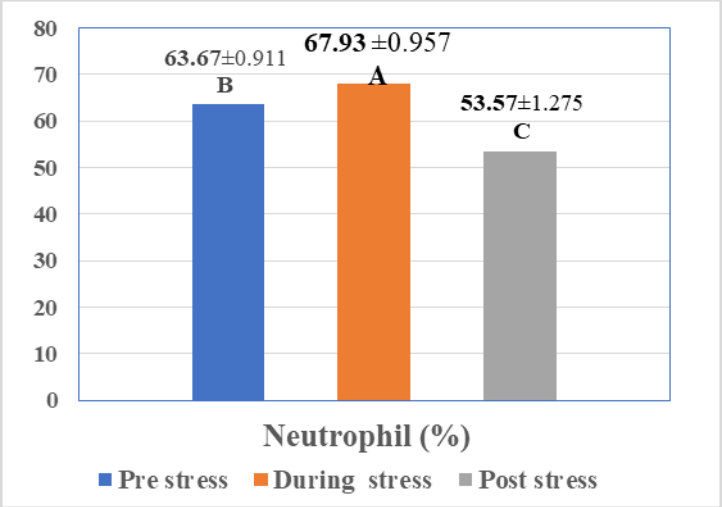

Figure 6: The effect of psychological stress this figure shows the between the Neutrophil(%). this figure shows the significant difference between the Neutrophil count in three groups of study, (B) non-significant difference between pre-stress and during stress , (A) significant difference between during stress and post stress, (C) significant difference between pre stress and post stress.

Table 2 represents the effect of psychological stress on RBC counts (cellx106/mm3) and hemoglobin (g/l) results. It shows that the RBC count significantly increases during stress and returns to decline after the cause of stress is diminished. On the other hand, the hemoglobin concentration also increases significantly during exposure to psychological stress and returns to decrease after the stress disappears as shows in figure 6 and 7.

Table 2: The effect of psychological stress on RBC counts (cellx10^6/mm^3) and hemoglobin (g/l) in male and female students at University

Parameter	Pre stress	During stress	Post stress	LSD
RBC (cellx10^6/mm^3)	**4.47** ±0.07 B	**5.03** ±0.11 A	**4.30** ±0.06 B	**0.25**
Hemoglobin (g/l)	**13.07** ±0.21 B	**14.74** ±0.50 A	**12.64** ±0.20 B	**1.02**

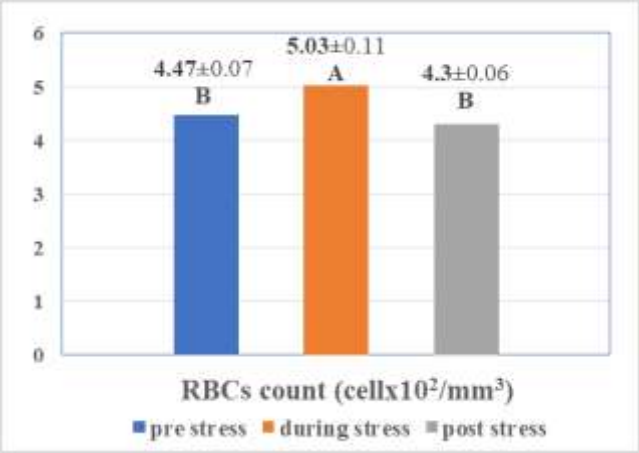

Figure 7 : The effect of psychological stress on Red Blood Cell. This figure shows the significant difference between the Red Blood Cell count in three groups of study, (A) significant difference between pre-stress and during stress , and significant difference between during stress and post stress, but (B) non-significant difference between pre-stress and post stress

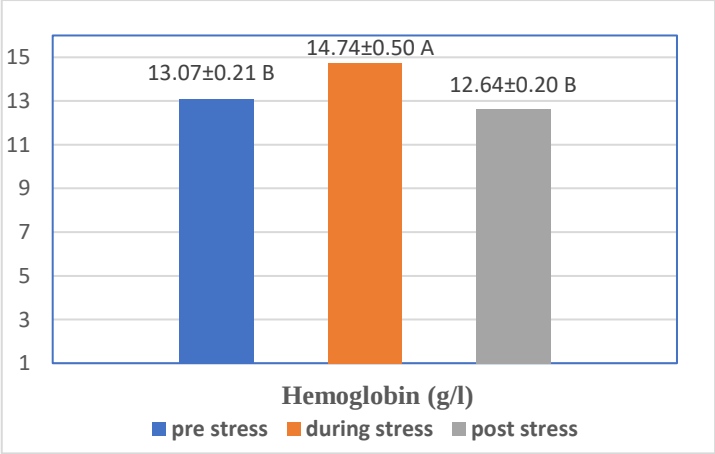

Figure 8: The effect of psychological stress on Hemoglobin. This figure shows the significant difference between the Hemoglobin in three groups of study, (A) significant difference between pre-stress and during stress, and significant difference between during stress and post-stress, but (B) non-significant difference between pre-stress and post stres

Table 3 represent the effect of psychological stress on cortisol (mcg/dl) results. It showed that the cortisol significantly increase during stress and return to decline after diminishing of stress cause, as shows figures 8 and 9.

Table 3: The effect of psychological stress on Cortisol (mcg/dl) in male and female students in University

Parameter	Pre stress	During stress	Post stress	LSD
Cortisol (**mcg/dl**)	**290.3** ±44.03 B	**458.2**±60.86 A	**200.1** ±31.70 B	137

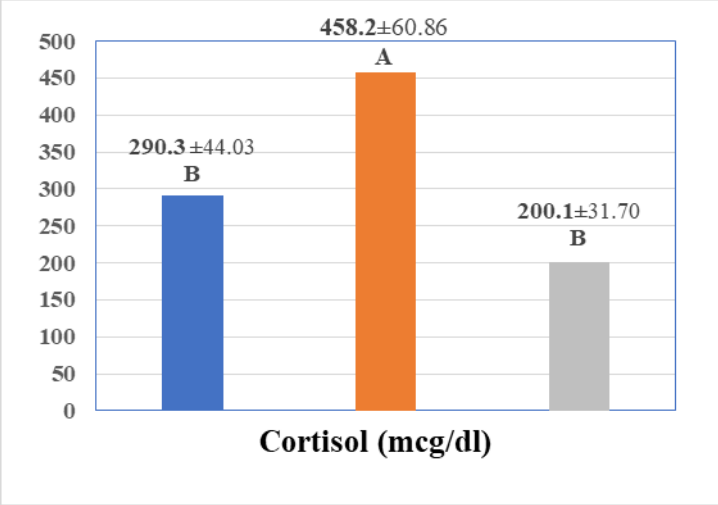

Figure 9: The effect of psychological stress on Cortisol (mcg/dl), This figure shows the significant difference between the Cortisol in three groups of study, (A) significant difference between pre-stress and during stress , and significant difference between during stress and post stress, but (B) non-significant difference between pre-stress and post stress.

4. Discussion

The result of the study indicated a significant decrease in White blood cell count during stress and a return to rise after diminishing of stress cause. On the other hand, the lymphocyte percentage also decreases significantly during exposure to psychological stress and returns to enhanced and elevated after the disappearance of stress. This result agrees with Sharma and Gupta (2015) White blood cell types showed significant changes, with monocytes, lymphocytes, eosinophils, and basophils showing the most striking variations. However, according to certain research, stress has no discernible effect on the number of white blood cells (WBCs). This result is consistent with other studies conducted at the University of Al-Qadisiya, which also found no statistically significant variations in total WBC counts (Al-Zamely, 2012).

Alhmoud (2021), in this study, indicated an increase in the WBC count during examination but a decrease after 72 hr of examination, while a decrease in the lymphocyte count during the examination. Chronic stress has been linked to decreased lymphocyte counts because it causes lymphocytes to redistribute and attach to the endothelium. In this study, Suleiman D. *et al*. (2015) showed that elevated neutrophil counts during prolonged stress correlated with elevated acute phase reactant levels and TNF-alpha functions. The Impact of Lentinan (β-glucan) Supplementation on Leukocyte Profiles in Intact Rabbits; when administered at a dosage of 10 mg/kg body weight, Lentinan (β-glucan) significantly increased the total leukocyte count in intact rabbits. Additionally, Lentinan had a significant impact on some subtypes of white blood cells, as seen by higher proportions of lymphocytes and monocytes and lower proportions of neutrophils and the neutrophil-to-lymphocyte ratio (NLR) (Alkenany and Khalil, 2022).

The current study shows a significantly increase in RBC count and hemoglobin during stress and returns to decline after the cause of stress is diminished, this agrees with study of Balgimbekov and Tashenova (2014) found that an increase in red blood cell activity is correlated with a positive shift in the prooxidant-oxidative balance in cell membranes, which is attributed to an increase in average red blood cell volume. which stated that the stress of academic examinations noticeably affects the erythrocyte counts. Shawesh *et al.*'s (2020) study, which shows that this study did not detect any evidence for a change in the hemoglobin content. Elevated the CBC values (WBC, Hb, and Plt) in the PCOS group compared to the control group (Abbas, *et al*., 2023).

The level of cortisol in current study shown increase during stress, This study agreement with study Maduka *et al*., (2015) which shows the association between adrenaline, cortisol, lipid profile, and blood glucose in undergraduate students during examination stress, therefore the level of cortisol is elevated in students during examination stress.

During the test, cortisol levels were raised, demonstrating the typical hormonal response to stress. Studies have demonstrated that the cortisol response to academic stress varies, ranging from no change to even lower levels (Takatsuji *et al*., 2008), to significantly higher levels (Maduka *et al*., 2015) as we shown in result of this study. Stress related to exams stimulates the HPA axis, which is why hormone levels are rising (Herman *et al*., 2016). While it came to perceived stress, there was a gender difference; while under stress, women scored higher than men. According to this belief, adult females are more likely than males to suffer from anxiety and depressive problems (Pfau *et al*., 2015).

5. Conclusion

There were clear evidence that the psychological stress can cause significant decrease in some immunological parameters like total leukocytes and lymphocytes count during psychological stress as compared to their values before exposing to stress. And it return to rise after demising the cause of stress. Also found the academic stress effect the concentration of stress hormone (cortisol). Recommendations of this study include study the effects of academic stress on the other hormones such as (thyroid hormones, sex hormones).

Funding: This research received no external funding.
Conflicts of Interest: The authors declare no conflict of interest.
Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers.

References

[1] Abbas, A. N., Al-Masaoodi, R. A., Hassan, S., & Al-Shemery, M. K. The relationship between hormonal levels and hematological parameters in cystic ovarian syndrome. *Journal of Medicine and Life*, 2023;*16*(6), 937.
[2] Adom, D., Chukwuere, J., & Osei, M. (2020). Academic stress among faculty and students in higher institutions. *Pertanika Journal of Social Sciences & Humanities*, 28(2).
[3] AL-azawi, T. SS; ALkenany, M. R. (2017) Complementary effect of vitamin C and Zinc on some blood cells and serum proteins related to immunity in intact and ovariectomized rabbits. *J Entomol Zool Stud*, 2017, 5.5: 1296-1303.
[4] Alhmoud, J. F., Farah, H. S., Al-Qaisi, T., Hamdan, W., Oriquat, G. A., Atoom, A., & AlQaisi, K. M. (2021). The Changes in Some Hematological Parameters among University Students Due to Stressful Conditions During and after Examinations Period. *Indian Journal of Forensic Medicine & Toxicology*, 15(1).
[5] Alkenany, M. R., & Khalil, L. W. (2022) The effect of Lentinan administration on some immunological and biochemical parameters in intact rabbits. *International Journal of Health Sciences*,2022; 6(S3), 6362–6381.
[6] Almis, B.H. and Aksoy, I. (2018) Mean Platelet Volume Levels in Patients with Generalized Anxiety Disorder. Psychiatry and Clinical Psychopharmacology, 28, 43.
[7] Al-Zamely, H. A. N. (2012). The effect of stress of academic examination on some physiological parameters of boy students of veterinary medicine College "university of AlQadisyiah. *Al-Qadisiyah Journal of Veterinary Medicine Sciences, 11(*1), 117-121.
[8] Asoglu, M., Aslan, M., Imre, O., Kivrak, Y., Akil, O., Savik, E., Buyukaslan, H., Fedal, U. and Altindag, A. (2016) Mean Trombosit Volume and Red Cell Distribution Width Levels in Initial Evaluation of Panic Disorder. Neuropsychiatric Disease and Treatment, 12, 2435-2438.
[9] Balgimbekov SA, Tashenova GK. (2014) Features of hematological parameters in students with different levels of motor activity in modern education. Bul Perm State Univ S Biol. 2014;2:57-60
[10] Banich, M.T., & Compton, R.J. (2018). Cognitive Neuroscience. Cambridge: Cambridge University Press
[11] Borgohain, B. and Boro, M. (2020) A Study on Mean Blood Platelet Volume Levels in Patients with Depression and Generalized Anxiety Disorder. *Journal of Medical Science and Clinical Research*, 8, 658-664.
[12] Dhama, K., Latheef, S. K., Dadar, M., Samad, H. A., Munjal, A., Khandia, R., ... & Joshi, S. K. (2019). Biomarkers in stress related diseases/disorders: diagnostic, prognostic, and therapeutic values. Frontiers in molecular biosciences, 6, 465402.
[13] Durmaz, T., Ozdemir, O., Keles, T., Bayram, N.A., Akcay, M., Yeter, E., Ayhan, H. and Bozkurt, E. (2009) Platelets and Sympathetic Activity in MI. *Turkey Journal of Medical Science*, 39, 259-265.
[14] Elenkor IJ and Chrousos E. (2002). Stress hormones, proinflammatory and anti-inflammatory cytokines and autoimmunity. Annals of Academic Science. 966: 290-303.
[15] Hellhammer, D.H., Wust, S. and Kudielka, B.M. (2009). Salivary Cortisol as a Biomarker in Stress Research. Psychoneuroendocrinology; 34:163-171.
[16] Herman JP, McKlveen JM, Ghosal S, Kopp B, Wulsin A, Makinson R, et al. (2016) Regulation of the Hypothalamic-Pituitary-Adrenocortical Stress Response. Compr Physiol. 2016; 6(2):603–21.
[17] Hettiarachchi M, Fonseka C, Gunasekara, P. Jayasinghe, D. Maduranga D. (2014) 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
[18] Kupriyanov, R. and Zhdanov, R. (2014). The eustress concept: Problems and out looks. World Journal of Medical Sciences, 11(2), 179-185.
[19] Lin, S. H. and Huang, Y. C. (2014). Life stress and academic burnout. Active Learning in Higher Education, 15(1), 77-90.
[20] Maduka IC, Neboh EE, and Ufelle SA (2015). The relationship between serum cortisol, adrenaline, blood glucose and lipid profile of undergraduate students under examination stress. African Health Science;15(1):131-6.
[21] Martinez, P.M., Martinz, L.R. and Ramos, R. (2009). Cortisol and Glucose: Reliable Indicators of Fish Stress. Pan *American Journal of Aquatic Sciences*, 4, 158-178.

[22] Ouda, S., Alaki, S., Safi, M. A., Nadhreen, A., & Johani, K. A. (2016). Salivary stress biomarkers–Are they predictors of academic assessment exams stress. J. Clin. Exp. Pathol, 15, 276-279.
[23] Pfau ML and Russo SJ. (2015) Peripheral and Central Mechanisms of Stress Resilience. Neurobiol Stress. 2015; 1:66–79.
[24] Sandman, C.A., Davis, E.P., Buss, C. and Glynn, L.M. (2011). Prenatal Programming of Human Neurological Function, *International Journal of Peptides*, 2011.
[25] Sarafino, E. P. and Smith, T. W. (2014). Health psycholology: Biopsychosocial interactions. USA: John Wiley & Sons.
[26] Sharma, N. and Gupta, V. (2015). Effect of examination stress on hematological and hemodynamic parameters in students. Int J Pharmacol Physiol., 1(1), 24-29.
[27] Shawesh, F., Khdeir, A., Alzaruge, R., Abu-Alqasim, M., Kharbash, A. and Elazomi, A. (2020). Effect of examination stress on blood cells parameters among medical technology students in the university of zawia, libya. *European journal of pharmaceutical and medical research*, 7(1), 171-176.
[28] Suleiman D, Abdullah A, Mahmut B, Aslıhan O, Mehmet G, Mehmet C, Özlem D, Aytekin S. (2015) Neutrophil-lymphocyte ratio in patients with major depressive disorder undergoing no pharmacological therapy. Neuropsychiatr Dis Treat. 2015; 27(11) :2253-8.
[29] Takatsuji K, Sugimoto Y, Ishizaki S, Ozaki Y, Matsuyama E and Yamaguchi Y. (2008) The effects of examination stress on salivary cortisol, immunoglobulin A, and chromogranin A in nursing students. Biomed Res. 2008; 29(4):221–4.