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Effect Of Examination Stress On Some Biochemical Indices In Tikrit College Students

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Abstract. This study aimed to investigate the physiological impact of academic stress, particularly examination stress, on biochemical indices among college students at the University of Tikrit. A total of 60 healthy students (30 in an experimental group and 30 in a control group) aged 19-21 years participated in this cross-sectional study. Salivary samples were collected during pre-examination and post-examination periods to measure levels of cortisol, malondialdehyde (MDA), glucose, and total antioxidant capacity (TAC). The results indicated significant increases in cortisol, MDA, and glucose levels in the pre-examination period compared to the post-examination period, suggesting heightened stress responses. Conversely, TAC levels were significantly higher in the post-examination period, indicating a potential recovery or response to oxidative stress. The findings support the hypothesis that academic examinations can induce significant physiological stress in students, reflected by changes in salivary biochemical markers. These stress responses may influence overall student health and academic performance, suggesting the need for strategies to manage stress among students during examinations.

Keywords: examination, biochemical, indices, Tikrit College and students.

INTRODUCTION

Stress was originally described by Selye as a general bodily reaction to any kind of demand for change. This definition has been expanded by other authors to mean a condition of imbalance or disturbed equilibrium, and when discussing the stress response, as a broad alert in a homeostatic system that triggers a widespread, nonspecific increase in neurological activity. More modern interpretations define stress as either an actual or perceived threat to one's physical and mental well-being, leading to a series of adaptive biological and behavioral changes[(1)].

Academic exams are known to significantly affect students' well-being. When a person is faced with a challenge or threat, the sympathetic adrenal medullary (SAM) system is activated, leading to increased release of epinephrine and norepinephrine. This activation prepares the body to handle stressful situations. If stress persists, it triggers the hypothalamo-pituitary-adrenal (HPA) axis, resulting in elevated levels of hormones such as adrenocorticotropic hormone (ACTH) and cortisol, which are involved in the body's stress response [(2)].

Cortisol, a steroid hormone, is synthesized in response to ACTH and is the final product of the hypothalamic-pituitary-adrenal (HPA) axis. It primarily influences gluconeogenesis, proteolysis, and lipolysis, and contributes to hyperglycemia. Additionally, cortisol affects

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immune and inflammatory responses, sexual and reproductive health, and is also involved in the regulation of bone metabolism [(3)].

Cortisol is commonly utilized in research as a reliable physiological indicator of stress, with its levels mirroring psychological stress. Under stressful situations, the adrenal cortex ramps up its production of cortisol. This hormone is also connected to a variety of psychobiological and physiological functions, including learning and memory, apoptosis, neural growth, metabolism, and immune response [(4)].

Oxidants, including hydrogen peroxide, protein peroxides, lipid hydroperoxides, and peroxynitrites, are produced under both normal and pathological conditions. Given its high oxygen consumption and relatively poor antioxidant defenses, the brain is particularly susceptible to oxidative damage. Continuous oxidative harm to the brain can trigger further depressive episodes in individuals already experiencing depression [(4)].

Aim of the study

The aim of this study is to evaluate the cortisol, malondialdehyde, and total antioxidant capacity in students pre and post examination .

Materials and methods

In this study, the experimental group comprised 30 healthy students 17 females (56.6%) and 13 males (43.33%) ranging in age from 19 to 21 years, with an average age of 20.5 ± 0.39 years. These students underwent comprehensive medical screenings and examinations prior to their participation. The control group also included 30 healthy students, with 20 females (56.7%) and 10 males (43.3%), who were similarly aged (average age 20.4 ± 0.5 years). Their medical examinations were conducted during breaks without the influence of any stress-inducing factors.

Setting and Design: It is a cross-sectional study.

Experimental design

The research was conducted at two distinct times: first, 1 or 2 days before the exams (pre-examination period), and second, 1 week after the final written exam during the holiday period as students awaited their results (post-examination period). The average age of the participants was 22 years.

Inclusion criteria

1. Participants aged between 19 to 21 years.
2. No exposure to stressors for at least two months before the study.
3. No consumption of medications or biologically active supplements for at least three months before the study.
4. No presence of acute or chronic physical illnesses.
5. Provision of a signed voluntary consent form agreeing to participate in the study.

Exclusion criteria:

1. Exposure to any stressors within two months before the study.
2. Use of any medications within three months before the study.
3. Presence of any acute physical diseases.
4. Chronic physical diseases in acute or sub/decompensated stages, or any infectious diseases.
5. Any type of cancer.
6. Any diagnosed mental disorders.
7. Alcohol or drug abuse.
8. Lack of consent to participate in the study.

The study protocol was approved by the Ethics Committee of Tikrit University of college of medicine, Iraq. Written informed consent was obtained from the students.

Collection of sample

Saliva samples were gathered between 9 and 11 a.m., following a protocol where participants first rinsed their mouths with distilled water for 5 minutes and then spit out unstimulated saliva for another 5 minutes in a calm setting. To eliminate squamous cells and other cellular debris, the saliva was then centrifuged at 3000 rpm for 10 minutes in an Eppendorf centrifuge set at 4°C.

Cortisol, and MDA were analyzed by ELISA kit. Serum glucose levels was measured by spectrophotometric

Statistical Analysis:

All data are reported as the mean plus or minus the standard deviation. The Student t-test was employed to analyze the data, with results deemed statistically significant if the p-value was less than 0.05.

Results

The current study shows that the average salivary cortisol level in the group before the examination (12.198 ± 0.914) was significantly higher compared to the group after the examination (9.59 ± 1.19), as illustrated in Figure 4.1.

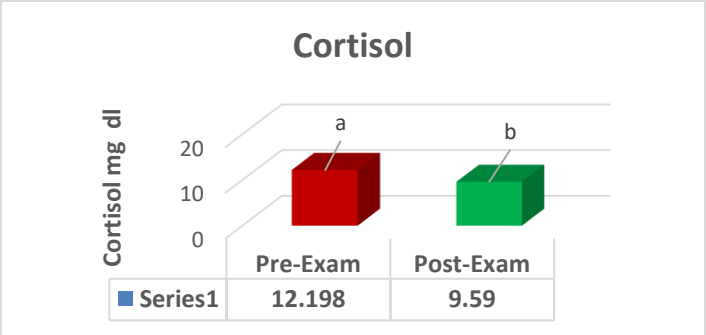

Figure4.1. Salivary levels of cortisol in study groups

This study demonstrated that the MDA was significantly higher at a P value of 0.0006 in pre examination group compared to post- examination group 4.319 ± 0.606 versus 2.640± 0.340 respectively, as shown in figure 4.2.

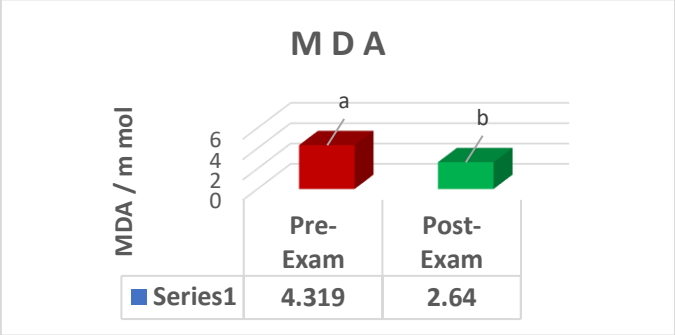

Figure 4.2. Salivary levels of MDA in study groups

The salivary glucose level in the pre examination group (4.824± 0.423) was significantly higher (*P*=0.0004) than the post examination group (3.910 ± 0.442) as shown in table (4.3).

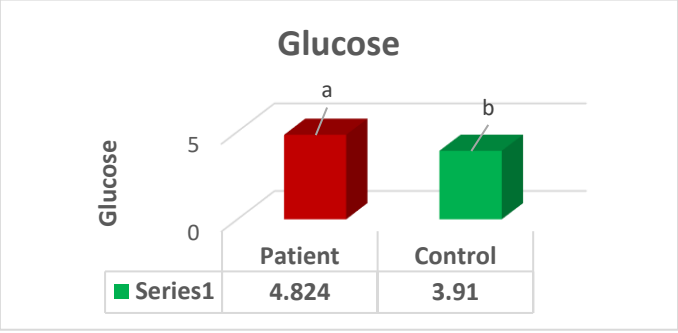

Figure 4.3. Salivary levels of glucose in study groups.

According to Figure.4.4., TAC of saliva in students was significantly higher in pre examination group (115.71± 7.61) compared to those with post examination group (162.33 ± 9.58); P= 0.0007, as evident in the following figure 4.4

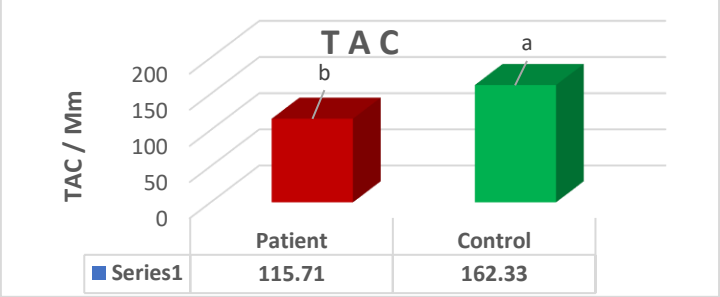

Figure 4.4. Salivary levels of TAC in study groups

Discussion

In the current analysis, it was observed that the levels of glucose in salivary samples of students were notably lower compared to those of the control group. This finding aligns with the research conducted by Jena et al [(3)]. Glucocorticoids act as functional antagonists to insulin, reducing insulin sensitivity and raising blood glucose levels through several mechanisms. These include: Increasing the production of crucial enzymes that drive gluconeogenesis, such as phosphoenolpyruvate carboxykinase (PEPCK) and glucose-6-phosphatase, (b) promoting proteolysis and lipolysis, which increase the levels of amino acids and glycerol used in gluconeogenesis, (c) reducing insulin production from the pancreas, and (d) blocking glucose transporter-4, which is responsible for glucose uptake into peripheral tissues. Additionally, catecholamines boost blood glucose levels by promoting the secretion of cortisol and glucagon and by increasing metabolic rate, glycogenolysis, and gluconeogenesis. This rise in blood glucose induced by glucocorticoids and catecholamines serves to fulfill the higher energy demands during stress [(3, 11,12)].

Stress from examinations triggers specific physiological responses in a student's body that are adaptive and result in alterations to hormonal balance [(13)].

Salivary total antioxidant capacity in the student group, the results of this study showed significantly lower levels of salivary TAC compared with those in the control group. This decrease might be regarded as a protectively active response to inflammation tissue shifts, which includes the enhancement of oxidative processes and a deficit of antioxidants to neutralize the surplus reactive oxygen species [(14-16)].

These results indications are consistent with the results of numerous previous studies. The findings of Piedade Sequeira and Naik indicated [17] that the total antioxidant capacity of saliva significantly decreased on the examination day compared to after the exam. Moreover, a pronounced negative correlation was found between stress scores and saliva TAC. Since then, the researchers have suggested that salivary TAC may serve as a good marker of mental stress. At the same time, they recommend evaluating the possibility of taking antioxidant therapies by students when the level of stress increases, for example, during exams.

Pani et al. [18] The study explored how regular physical activity influences psychological stress levels and salivary total antioxidant capacity (TAC). Despite no significant variances in self-reported exam stress levels, individuals who engaged in regular physical activity experienced a less substantial reduction in TAC. This indicates that regular physical exercise could help safeguard students from the oxidative stress related to academic demands. Furthermore, the study revealed that salivary malondialdehyde (MDA) levels were significantly higher ($p < 0.05$) in the pre-exam group compared to the post-exam group.

In consistence with our findings Cernak et al. (2000) [19]. reported that young volunteers subjected to chronic or subchronic stress for over three months exhibited elevated levels of malondialdehyde, a marker of oxidative damage to lipids. Additionally, they observed an increase in superoxide production in the plasma and alterations in antioxidant defenses in these individuals. These changes collectively suggest that psychological stress is associated with heightened oxidative stress.

Lesgards et al. (2002) [20] The research examined how various lifestyle factors affected the resilience of red blood cells to oxidative challenges. Findings from this study highlighted that psychological stress significantly impacts the antioxidant status.

The exact mechanisms by which oxidative stress markers operate remain uncertain. A reduction in antioxidant capacity might stem from the depletion of antioxidants due to oxidative stress. A possible reason for the heightened oxidative stress observed during psychological stress may be the increased levels of catecholamines in the bloodstream, which are known to produce reactive oxygen species [21].

The main finding of this study was that pre examination group have significantly higher salivary cortisol levels than post examination group because the stress influences the activity of the hypothalamic pituitary adrenal axis and consequently increase the serum cortisol level

The results of this study align with previous research findings. Joseph C. Awalu [22] noted an increase in stress and anxiety among first-year medical students, attributing it to academic pressure. Ganesh Pradhan and colleagues [23] found that exams, as stressors, negatively affect the cognitive functions of first-year medical students. Krzysztol [24] observed elevated cortisol levels in response to academic and laboratory stressors. In contrast, Patricia et al. [25] reported decreased cortisol levels during exam stress, and Larson et al. [26] found no significant changes in plasma cortisol levels before, during, or after exams.

Conclusion

1. The findings of this study confirm that examinations are a stressful experience that impacts both mental and physical health. Elevated blood sugar levels above baseline may serve as a marker of stress. Therefore, monitoring glucose levels can be beneficial for informing students about the negative effects of stress and for providing necessary guidance on stress management strategies.
2. The increase in TAC level in students is an indication of oxidative stress.
3. The physiological impact of stress from academic exams, such as heightened cortisol secretion, may be a temporary condition, as normal bodily equilibrium typically resumes once the stressor is removed. Nevertheless, the ongoing pressure associated with exams can make them a source of dread.

Recommendations

1. While a link was observed between academic performance and exam stress within the semester system, a broader study involving multiple institutions across Iraq is necessary to rule out any socioeconomic influences, considering Iraq's status as a developing country.
2. Additional longitudinal research is needed to explore the impact of oxidative stress caused by examinations on university students.

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