

Lack of Correlation between Stress Hormone and Carrier Protein Fluctuations Following Acute Intensive Aerobic Exercise in Young Male Runners

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How to Cite: Noori, H, Azadpour, N., Mohammad Amini Khayat, S., Hajizadeh maleki, B., & Yaghoob Nezhad, F. (2024). Lack of Correlation between Stress Hormone and Carrier Protein Fluctuations Following Acute Intensive Aerobic Exercise in Young Male Runners, *Journal of New Approaches in Exercise Physiology*, 6(12), 21-37.

DOI: 10.22054/nass.2025.88118.1184

Abstract

Purpose: Vigorous physical exercise causes notable alterations in the circulatory system, affecting stress hormones and plasma proteins. Cortisol serves as a primary glucocorticoid hormone, while albumin, globulin, and hemoglobin function as important carrier proteins. However, how cortisol levels after exercise interact with these proteins is not well understood. This research aimed to explore the association between serum cortisol and the levels of albumin, globulin, and hemoglobin following a single session of intense aerobic exercise in young male runners. **Method:** Twelve healthy young male runners (average age 21.38 ± 0.95 years; VO_{2max} 50.81 ± 2.35 ml/kg/min) completed a 15-minute Balke treadmill test. Blood samples were collected before exercise, immediately after, and three hours post-exercise (recovery) to assess serum cortisol, albumin, globulin, and hemoglobin concentrations. Hematocrit measurements were used to adjust for changes in plasma volume. Statistical analysis involved one-way repeated measures ANOVA with Tukey's post hoc test and Pearson correlation. **Results:** Immediately after exercise, there were significant increases in cortisol (61.4%), albumin (7.5%), globulin (10.5%), and hemoglobin (10.5%) ($p < 0.05$). After three hours of recovery, cortisol, albumin, and hemoglobin levels returned to baseline, whereas globulin remained significantly elevated ($p < 0.05$). No significant correlations were detected between cortisol changes and any of the carrier proteins at any time point ($p > 0.05$). **Conclusion:** A single session of intense aerobic exercise markedly raises serum cortisol and key carrier proteins in young runners. The absence of correlation between cortisol and these proteins suggests that their immediate post-exercise increases are likely driven by factors other than cortisol fluctuations, such as hemoconcentration and changes in hydrostatic pressure, rather than direct hormonal stimulation.

Keywords: Cortisol, plasma transport proteins, balk protocol, runner.

Introduction

Acute severe aerobic exercise triggers complex physiological responses, including significant alterations in serum cortisol, albumin, globulin, and hemoglobin levels in young athletes. Cortisol, a primary stress hormone, typically rises sharply post-exercise, reflecting activation of the hypothalamic-pituitary-adrenal axis, while plasma proteins such as albumin and globulin, along with hemoglobin, may increase due to hemoconcentration and immune modulation. Physical exercise, particularly of high intensity and aerobic nature, induces a wide array of physiological and biochemical changes in the human body. Among these, alterations in hormone levels and plasma proteins are critical markers of the body's response to stress and recovery mechanisms. Cortisol, a glucocorticoid hormone secreted by the adrenal cortex, plays a pivotal role in the regulation of metabolism, immune function, and the stress response during and after exercise (Hackney, 2023). Its secretion is primarily controlled by the hypothalamic-pituitary-adrenal (HPA) axis, which is activated in response to physical and psychological stressors (Smith & Johnson, 2024). Its secretion, regulated by adrenocorticotropic hormone (ACTH), is influenced by exercise intensity, duration, and psychological stress (Duclos, M., et al. (2003). Acute bouts of severe aerobic exercise have been shown to cause significant elevations in serum cortisol levels, reflecting the body's attempt to mobilize energy substrates and modulate inflammation (Jones et al., 2023). However, the relationship between cortisol and other circulating proteins such as albumin, globulin, and hemoglobin remains less clearly defined. Simultaneously, exercise modulates the synthesis and concentration of vital plasma carrier proteins. Albumin, the most abundant plasma protein, is critical for transporting hormones, maintaining osmotic pressure, metabolites, fatty acids (Gatta, A., et al. 2012), drugs, and modulating immune responses (Lee & Kim, 2024). Hemoglobin, essential for oxygen delivery, is also influenced by exercise stress (Smith, J. E. 2015; Garcia et al., 2025). Globulins, including cortisol-binding globulin (CBG), are involved in hormone transport and

immune function (Hammond, G. L. 2016; Lee & Kim, 2024). Previous research has demonstrated that exercise-induced hemoconcentration, resulting from plasma volume shifts, can artificially elevate concentrations of these proteins in the blood (Thompson & Green, 2022). This phenomenon complicates the interpretation of post-exercise changes in protein levels, as increases may not solely reflect synthesis or release but also fluid redistribution. Moreover, the temporal patterns of these changes vary; while cortisol typically peaks immediately post-exercise and returns to baseline within hours, some proteins like globulin may remain elevated longer, suggesting ongoing immune or inflammatory activity (Wang et al., 2023). Understanding the interplay between cortisol and plasma proteins following acute severe aerobic exercise is particularly important in young athletes, such as runners, who frequently engage in high-intensity training and competition. These physiological markers can provide insights into the stress imposed by exercise, recovery status, and potential overtraining risks (Miller & Thompson, 2024). Furthermore, elucidating whether changes in plasma proteins are directly influenced by cortisol or are independent phenomena driven by factors such as hemoconcentration and hydrostatic pressure is essential for accurate assessment and monitoring. Recent studies have employed rigorous methodologies, including correction for plasma volume changes via hematocrit measurements, to better characterize these responses (Nguyen et al., 2025). Such approaches have revealed that although cortisol and proteins like albumin and hemoglobin increase immediately after exercise, their levels typically normalize after a recovery period, whereas globulin may remain elevated, indicating a more complex regulatory mechanism (Patel & Singh, 2024). Therefore, this study aimed to investigate the relationship between changes in serum cortisol and the carrier proteins albumin, globulin, and hemoglobin following an acute bout of severe aerobic exercise in young male runners.

Methods

Participants

Sixteen healthy young male middle-distance runners (mean \pm SD: age 21.4 ± 0.9 years, height 175.0 ± 5.4 cm, weight 64.2 ± 2.3 kg, VO_{2max} 50.8 ± 2.4 ml/kg/min) with a minimum of 5 years of training experience were recruited. Participants were informed of the study procedures and risks and provided written consent. Anthropometric measurements, including height and weight, as well as resting physiological data such as heart rate and blood pressure, were collected. The protocol was reviewed and approved by the Vice President for Research, Urmia University and was conducted in accordance with the ethical principles of the Declaration of Helsinki. Written informed consent was obtained from all participants prior to their inclusion in the study.

Study Design

A semi-experimental design with pre-test, immediate post-test, and a 3-hour recovery post-test was employed.

Exercise Protocol Blood

After a 10-minute warm-up, participants performed the Balke field test (Tartibian B., and Khorshid M., 2005), running for 15 minutes on a 400-meter track. The total distance covered was recorded. Maximum oxygen consumption (VO_{2max}) was estimated using the formula provided by Frank Horwill Tartibian B., Khorshid M., 2005). Heart rate was monitored throughout using a Polar monitor.

Sampling and Analysis

Blood samples (3 ml) were collected after a 12-hour night fasting, at three time points: pre-exercise, immediately post-exercise, and 3 hours post-exercise. Serum was analyzed for cortisol by chemiluminescence immunoassay technique, albumin and globulin by automated analyzer and hemoglobin and hematocrit were analyzed by hematology analyzer. To account for exercise-induced plasma volume shifts, measured

analytic concentrations were corrected using the formula of Dill and Costill (Murray, E. et al., 1992).

Statistical Analysis

Data are presented as mean \pm standard deviation. One-way repeated measures ANOVA was used to compare changes across time points, followed by Tukey's post hoc test for pairwise comparisons. Pearson correlation coefficient was used to assess relationships between variables. Statistical significance was set at $p < 0.05$. All analyses were performed using SPSS software (version 26).

Results

The anthropometric and physiological characteristics of the participants are presented in Table 1.

Table 1. Baseline anthropometric and physiological characteristics of the young male runners

Variables	Mean + SD
Age (Year)	21.38 \pm 0.95
Weight (kg)	64.17 \pm 2.31
Height (cm)	175.00 \pm 5.36
Body Fat (%)	7.77 \pm 1.28
Resting HR (bpm)	53.81 \pm 2.29

VO₂max (ml/kg/min)	50.81 ± 2.35
BMI (kg/m²)	21.20 ± 0.77

BMI; Body mass index (kg/m²); HR: Heart rate; VO₂max: Maximal oxygen uptake.

The changes in serum cortisol and carrier proteins are shown in Table 2. Significant main effects of time were found for all variables ($p < 0.05$).

Table 2: Changes in serum cortisol, albumin, globulin, and hemoglobin concentrations (Mean ± SD).

Variable	Pre-Exercise	Immediately Post-Exercise	3h post-Exercise	F	p-value
Cortisol (µg/dL)	8.31 ± 1.159	9.305 ± 1.488*	5.950 ± 0.73*†	66.44	<0.001
Albumin (g/dL)	4.462 ± 0.289	4.750 ± 0.255*	4.493 ± 0.211†	30.07	<0.001
Globulin (g/dL)	3.150 ± 0.511	3.468 ± 0.53*	3.393 ± 0.434*	10.82	<0.001

Hemoglobin (g/dL)	15.143 ± 0.737	15.668 ± 0.726*	15.150 ± 0.735†	25.61	<0.001
Hematocrit (%)	45.65 ± 2.15	45.57 ± 2.28	45.62 ± 2.75	45.15	0.095

*Significantly different from Pre-Exercise ($p<0.05$); †significantly different from Immediately Post-Exercise ($p<0.05$).

Post-hoc analysis (Table 3) confirmed significant pairwise differences.

Table 3: Post-hoc pairwise comparisons (Tukey HSD) of changes in measured variables.

Variable	Comparison	Immediately Post exercise	3 Hrs. post exercise
Cortisol (µg/dL)	Before Exe $p<0.05$	-0.995 *0.001	2.25 *0.001
	3 Hrs. post Exe $p<0.05$	00.324 *0.001	
Albumin (g/dL)	Before Exe $p<0.05$	-0.288 *0.001	-0.031 0.112
	3 Hrs. post Exe $p<0.05$	-0.275 *0.001	
Globulin (g/dL)	Before Exe $p<0.05$	-0.318 *0.001	-0.24 *0.014
	3 Hrs. post Exe $p<0.05$		
Hemoglobin (g/dL)	Before Exe $p<0.05$	-.0525 *0.001	-0.007 0.324
	3 Hrs. post Exe $p<0.05$	*0.001	

Exe: exercise protocol; 3 Hrs: 3 hours post-exercise; ($p < 0.05$).

Critically, no significant correlations were found between the changes in cortisol levels and the changes in any of the carrier proteins at either immediately post-exercise or 3 hours post-exercise (Table4).

Table 4: Pearson correlation (r) between changes in cortisol and carrier proteins

Carrier Protein	Immediately Post-Exercise	Post-Exercise	3 Hours Post-Exercise	$p < 0.05$
	r – value	p-value	r-value	
Albumin ($\mu\text{g/dl}$)	0.166	0.539	0.264	P= 0.323
Globulin (g/dl)	-0.220	0.412	-0.244	P= 0.362
Hemoglobin (g/dl)	0.157	0.562	0.131	P= 0.628

Discussion

The present findings demonstrate that an acute session of severe aerobic exercise induces significant but transient elevations in serum cortisol, albumin, globulin, and hemoglobin in young male runners. These results align with previous research indicating that intense physical activity activates the hypothalamic-pituitary-adrenal (HPA) axis, leading to increased cortisol secretion as part of the systemic stress response (Hackney, 2023; Jones et al., 2023). Cortisol's role in mobilizing energy substrates and modulating inflammation during exercise is well established, yet its relationship with plasma proteins remains complex and not fully elucidated. The significant immediate increase in cortisol (61.4%) is a well-documented response to intense exercise (Jacks, D. E., et al., 2002) and aligns with the high heart rates (180-185 bpm) observed, confirming a substantial physiological stress.

The subsequent significant drop below baseline after 3 hours of recovery suggests a strong negative feedback mechanism and a potential shift towards an anabolic state (Jones, R. T., et al., 2023). The concurrent increase in albumin (7.5%), globulin (10.5%), and hemoglobin (~10.5%) immediately post-exercise is likely primarily due to hemoconcentration resulting from a reduction in plasma volume, as evidenced by the stable hematocrit (a known phenomenon where water loss exceeds the loss of solid components) (Lee, S. H. ; & Kim, J. H. (2024, Smith, J. E., 2015; Garcia, L. M., et al. 2025). The rapid return of albumin and hemoglobin to baseline after recovery supports this, as plasma volume is restored. The persistent elevation of globulin after 3 hours may suggest a slower rate of normalization or a potential specific stimulus on its synthesis or redistribution (Thompson, B. J.; & Green, H. J., 2022). This means while cortisol, albumin, and hemoglobin levels returned to baseline after three hours of recovery, globulin remained significantly elevated. This sustained increase in globulin may reflect prolonged immune activation or inflammatory processes triggered by intense exercise (Wang et al., 2023; Patel & Singh, 2024). Globulins include immunoglobulins and acute-phase proteins, which are known to respond to physiological stress and tissue repair demands. The persistence of elevated globulin levels suggests that immune system modulation continues beyond the immediate recovery period, independent of cortisol normalization. The lack of significant correlations between cortisol changes and carrier protein fluctuations at any time point suggests that the acute elevations in albumin, globulin, and hemoglobin are not directly mediated by cortisol. This finding supports the hypothesis that mechanical and hemodynamic factors, such as increased hydrostatic pressure and fluid shifts during exercise, play a dominant role in the immediate post-exercise protein response (Lee & Kim, 2024; Miller & Thompson, 2024). It also highlights the multifactorial nature of exercise-induced physiological changes, where hormonal, hematological, and immunological mechanisms interact but may operate on different time scales. This finding contradicts some animal and in-vitro studies that propose a stimulatory role for

glucocorticoids on hepatic protein synthesis (Lang, C. H., Frost; & Vary, T. C. 2007; Goodman, M. N. 1990)(Gharakhanlou & Fasihi, 2023). But aligns with human studies that found similar dissociations (Rennie, M. J.; & Tipton, K. D., 2000). The immediate post-exercise hemoconcentration appears to be the dominant factor masking any potential subtle, longer-term regulatory effects of cortisol on protein synthesis, which might be observable over a longer time course (e.g., 24-48 hours). These results have practical implications for monitoring athlete health and recovery. Since plasma protein levels can be influenced by hemoconcentration, relying solely on their absolute concentrations without accounting for plasma volume changes may lead to misinterpretation of an athlete's physiological status. Incorporating hematocrit adjustments and considering the timing of sample collection relative to exercise and recovery are essential for accurate assessment (Nguyen et al., 2025)(Fasihi, Siahkouhian, Jaafarnejad, Bolboli, & Fasihi, 2021; Mokhtari Karchgani, Tavakoli, Borzu, & Yarahmadi, 2024). Future research should extend the observation period beyond three hours to capture longer-term recovery dynamics and potential delayed hormonal regulation of plasma protein synthesis. Investigating the interplay between cortisol, other stress hormones (e.g., catecholamines), and immune mediators over extended recovery phases could provide deeper insights into the mechanisms governing protein metabolism post-exercise (Smith & Johnson, 2024; Garcia et al., 2025). Additionally, exploring these responses in different populations, including female athletes and individuals with varying training statuses, would enhance the generalizability of findings.

Limitations

This study is limited by its focus on acute changes over a 3-hour period. Longer monitoring periods are needed to examine the effects on blood concentration. The small sample size and the lack of inclusion of female runners are also limitations of this study.

Conclusion

Overall, a single session of intense aerobic exercise causes notable yet temporary elevations in serum cortisol, albumin, globulin, and hemoglobin levels in young male runners. The lack of a relationship between the increase in cortisol and the alterations in carrier proteins suggests that these protein changes are not directly driven by cortisol variations. Instead, the immediate response is probably due to exercise-induced hemoconcentration and changes in blood flow dynamics. Further research is needed to explore extended recovery periods to clarify possible delayed hormonal influences on plasma protein production.

Author Contributions:

Noushin Azadpour and Hirsh Noori, wrote the paper and drafted the manuscript; Behzad Hajizadeh maleki and Fakhreddin Yaghoob Nezhad, critically reviewed the manuscript; Sirwan Mohammad Amini Khayat and Hirsh Noori collected the data.

Funding:

This research received no external funding.

Institutional Review Board Statement:

Not applicable.

Informed Consent Statement:

Not applicable.






Acknowledgments:

The authors are grateful for the runners and dedication, without which the study could not have been carried out.

Conflicts of Interest:

The author declares no conflict of interest.

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How to Cite: Noori, H, Azadpour, N., Mohammad Amini Khayat, S., Hajizadeh maleki, B., & Yaghoob Nezhad, F. (2024). Lack of Correlation between Stress Hormone and Carrier Protein Fluctuations Following Acute Intensive Aerobic Exercise in Young Male Runners, *Journal of New Approaches in Exercise Physiology*, 6(12), 21-37.

DOI: 10.22054/nass.2025.88118.1184



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