



تحليل المؤشرات الحيوية في الدم (الكورتيزول، التستوستيرون، كرياتين كايينيز) خلال مراحل التحميل التدريبي والمنافسة لدى المصارعين

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تاريخ استلام البحث: ٢٠٢٥/ ٩ / ١

تاريخ قبول البحث : ٢٠٢٥/ ٩ / ١٤

الكلمات المفتاحية : الكورتيزول، التستوستيرون، كرياتين كايينيز، مراحل التدريب، إجهاد المنافسة
مستخلص البحث :

مستويات الكورتيزول ارتفعت بشكل كبير بعد التدريب قيمة ($p = 0.004$) ، مما يشير إلى زيادة في مستوى الإجهاد البدني. بالإضافة إلى ذلك، تم تخفيض مستويات التستوستيرون بشكل كبير بعد التمرين قيمة ($p = 0.032$)، مما يدل على حالة هدم مؤقتة. كما زادت مستويات كرياتين كايينيز بشكل ملحوظ بعد كل من التدريب والمنافسة قيمة ($p = 0.0001$) ، مما يشير إلى حدوث تلف عضلي كبير. **الاستنتاج:** تسلط هذه الدراسة الضوء على أهمية فترات الاستشفاء الكافية بعد التدريب والمنافسة على مستوى عالٍ لتجنب الإفراط في التدريب وتعزيز الأداء البدني. كما تم مناقشة كيفية أن المراقبة اليومية للمؤشرات الحيوية يمكن أن تساعد في تعديل الأحمال التدريبية وتحسين استراتيجيات التعافي.

أن تقييم ما إذا كانت المراحل المختلفة في التدريب والمنافسة تؤثر على مستويات الكورتيزول، التستوستيرون، وكرياتين كايينيز في المصارعين المحترفين. تم جمع عشرة بيانات من رياضيين محترفين في نادي الكاظمية، مع قياس المؤشرات الحيوية في أربع مراحل مختلفة مثل قبل التدريب، بعد التدريب، قبل المنافسة وبعد المنافسة. **الطريقة:** تم تنفيذ بروتوكول تدريبي موحد شمل تدريب المقاومة والتكيف القلبي الوعائي، مع التركيز على تطوير القوة وزيادة الحجم العضلي. تم تنفيذ تدريبات (HIIT) وتمارين قوة باستخدام وزن الجسم لتحسين اللياقة البدنية العامة. كما تم جمع عينات دم في كل مرحلة لاستخدام أساليب متقدمة لقياس مستويات الكورتيزول، التستوستيرون و كرياتين كايينيز. **النتائج:** لوحظ أن



competition were found ($p = 0.0001$), suggesting extensive muscle damage. These results highlight the importance of adequate recovery time post high intensity training and competition to avoid overtraining and enhance physical performance. It also explores the capacity for daily monitoring of these biomarkers to guide changes in training loads and recovery strategies.

Keyword ; Cortisol, Testosterone, Creatine Kinase (CK), Training Phases, Competition Stress.

Introduction

The sport of wrestling is very physically demanding, and athletes are required to have a great deal of strength and endurance in order to reach their potentials. Competitive preparation involves multiple phases of physical states and a wide selection of training practices ranging from high-intensity workouts to recovery methods, each phase directly affecting the peak performance capabilities for the athlete. In this sense, it is necessary to balance the organization of training in order to avoid excessive fatigue and overload as factors of interference that can harm performance. Understanding the physiological changes in biomarkers is therefore key in assessing training load effectiveness and hence maximizing advancement with training (Haller et al., 2023; Howlett, 1987; McGuigan et al., 2021).

Analysis of Blood Biomarkers (Cortisol, Testosterone, Creatine kinase) during Training Load and Competition Phases in Wrestlers

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Abstract

the effect of different stages of training and competition on cortisol, testosterone, and creatine kinase (CK) in professional wrestlers which have been followed up through measurements of those parameters. 10 players were taken from Al-Kadhimiya Club for wrestling. Biomarker measurements were obtained at four time points: pre training, post training, pre competition, and post competition. A standardized training program was followed involving resistance and cardiovascular exercise with an emphasis on strength and hypertrophy. HIIT (high intensity interval training) and body weight strength was added for total fitness. Blood sampling was carried out at each phase in order to implement sophisticated measurements of cortisol, testosterone and CK. Cortisol was significantly elevated after training ($p = 0.004$), indicating increased physical stress. In addition, testosterone concentrations were significantly lower after exercise ($p = 0.032$), indicating a state of temporary catabolism. Significant increases in CK levels following training and



based on showing the effect of different phases for training and competition processes in professional wrestlers on these biomarkers. This study investigates the association of performance decrements with temporal changes in training and competition phases.

This research is of great importance since it provides a thorough examination of variations in biomarkers at different stages of the year (during training and competitions) which helps to improve training strategies for wrestlers. This will help develop training programs that can be verified to prevent overtraining and injury by knowing exactly how the body responds to physical stress. Additionally, this work provides real data to design specific physiological-based training strategies useful for athletic performance without affecting health-related risks (Ammar et al., 2016; Nogueira et al., 2019; Taipale et al., 2018).

The objective of the study is to investigate cortisol, testosterone, and creatine kinase levels during a systematic non-specific resistance training method and different phases of wrestling loading and competition processes. Moreover, we will evaluate the effect of the high training loads on these biomarkers in professional wrestlers and quantify them not only during the training phase but also after competition. The hypotheses tested in this research were that cortisol levels will rise due to physical load during the training period, testosterone will decrease by long-term exertion, and creatine kinase will be up-regulated within a rich level of intense practice

Cortisol, testosterone, and creatine kinase (CK) are the most important parameters of biomarkers that have been applied for assessing the physical response to training in wrestlers. Cortisol is related to both physical and psychological stress and used as an important marker for detecting the influence of training load in athletes (Cevada et al., 2014a; Hloogeveen & Zonderland, 1996) Testosterone, one of the important anabolic hormones, significantly contributes to muscle development and regeneration, and it is also a hallmark of training in terms of how much muscle strength has been improved (Cevada et al., 2014b; Hayes et al., 2015; Taipale et al., 2018). One of those enzymes includes creatine kinase, an enzyme that can be used as a marker for muscle tissue damage as it significantly increases with high levels of physical activity and intense exercise, which offers a very valid tool to study the impact of training on muscular tissue (Halsen, 2014; Koch et al., 2014; Nogueira et al., 2019).

The importance of combat sports has been acknowledged for some time, especially in terms to the effects upon biomarkers and other physiological responses of athletes, although most work conducted so far had been focused around team sports or general training methodologies. Only a few study performed an analysis with the biomarkers of professional wrestlers, who faced different challenges because their muscular strength, resistance and flexibility were maintained by high-volume training (Bolach et al., 2016; Howlett, 1987). This lack of research setting justifies a study



experience in professional wrestling (compete independently regionally/nationally). In addition, the athletes have to be free of chronic diseases and physical ailments which could affect the results. We will not include wrestlers currently on performance-enhancing drugs (or have had any operations in the past six months) All participants will receive detailed information about the goals of the research, protocols, risks and will provide an informed written consent before study. The methodology will also choose a size appropriate sample pooling wrestlers with similar experience and training loads to provide consistency with length of training (i.e. reduce any confounding variables that would challenge the validity of the study as it relates to an individual's experience, training expertise, or general performance). Table 1 show the Descriptive characteristics for the Variables, mean (SD), for (age, Height, Weight and Experience)

Table 1: Descriptive Statistics for the Sample

Variables	Mean	Standard Deviation
Age (years)	22.6	1.35
Height (cm)	179.3	2.98
Weight (kg)	87.5	3.03
Experience (years)	4.8	1.32

and competition, leading to muscle tissue injury which is metabolic stress (Bolach et al., 2016; Koch et al., 2014; Lee et al., 2017; Rietjens et al., 2005).

Materials and Methods

Study Design

This study will employ a longitudinal, observational design to assess how cortisol, testosterone, and creatine kinase (CK) levels change in professional wrestlers during different phases of training and competition. The research will track ten male professional wrestlers from Al-Kadhimiya Club, aged 20 to 24 years. The study will span six weeks, with specific phases of training and competition. Study period The study will be performed between the dates of February 1, 2025 and March 15, 2025. Athletes will be assessed at three unique time points: pre-training load (before any substantial training or competition), pre-competition load, and post-competition. By examining changes over time, this protocol will provide a holistic view on how these biomarkers are affected throughout the various stages of training and competition in professional athletes. The selected biomarkers cortisone, testosterone and CK are widely used to evaluate stress, muscle recovery and muscle damage. These markers are used to evaluate the physiological stress and recovery processes in high-intense sport athletes (e.g. wrestlers).

Participants

The research will involve 10 male professional wrestlers meeting particular inclusion standards. Entrants must be aged between 20 and 24 years, with three or more years of



Procedure

The study will utilize a uniform training protocol that includes combined resistance and cardiovascular training. The resistance training block will focus on strength and hypertrophy, which will include lifts such as squats, deadlift, benches, and pull ups to effectively work the muscles used in wrestling's major muscle group. The cardiovascular portion will include HIIT and agility drills, wrestling specific. Athletes will train an average of five days per week training, with each session lasting from 90 minutes to two hours. The volume for each session will be adjusted according to the workload expressed as percentage of one-rep max (1RM) for strength exercises, and target heart rate zones for cardiovascular activities.

Blood-samples will be collected at three relevant time- points: before (baseline), after the training-load phase (just before competition), and immediately post-competition. In order to reduce the influence of diurnal variation in hormone concentrations, blood samples will be collected in the morning pre-prandially. All blood collection will be performed by certified medical professionals following standard procedures to ensure the safety and consistency of the samples. The timeline for each phase of the study will span over a period of six weeks, with two weeks allocated to the baseline and training load phases, followed by one week dedicated to competition preparation and post-competition analysis. Table 2 shown the Training Protocol of the study.

Data Collection

the biomarkers will be cortisol, testosterone, and creatine kinase (CK) because they are commonly referred to as being: stress marker; muscle recovery marker; and muscle damagemarker respectively. Cortisol and testosterone concentrations in blood samples will be determined by enzyme-linked immunosorbent assays (ELISAs) which is a well-validated and reliable technique for hormone quantification. ELISA was chosen to determine the sensitivity, specificity and reproducibility of this method because nothing is detectable in human serum at low concentrations of these hormones. Creatine kinase will be measured by an enzymatic assay, which is the standard method for the detection of muscle damage and widely used in both clinical and sports science research.

The blood samples will be processed immediately and stored at -80°C until they are ready for analysis. This storage method ensures that the biomarkers remain stable and do not degrade prior to testing. All assays will be carried out in a blinded manner to ensure impartiality in the results. Each participant's samples will be analyzed in duplicate to ensure reliability and reduce potential errors.



Table 2: show the Training Protocol

Phase	Duration	Primary Focus	Details
Baseline	2 weeks	Initial assessment and adaptation to exercises	- Collection of baseline biomarker data (cortisol, testosterone, CK) through blood samples at the start of the study. - Low-intensity training to assess physical performance before the onset of intensive training. - Body adaptation to physical effort without major changes in exercises.
Training Load Phase	2 weeks	Strength and hypertrophy development and cardiovascular conditioning	- Resistance Training: 4 days/week (exercises such as squats, deadlifts, bench presses, pull-ups) - Cardiovascular Conditioning: 2 days/week (HIIT, agility drills) - Intensity based on 1RM for resistance exercises and target heart rate zones for cardiovascular exercises.
Competition Preparation	1 week	Intensive training before competition	Focus on maintaining strength while enhancing sport-specific skills and agility drills. - Moderate intensity resistance training 2 days/week, focusing on wrestling-specific movement patterns.
Post-Competition	1 week	Recovery and analysis	Light training or rest with focus on recovery. Blood samples are collected after the competition for analysis.

Statistical Analysis

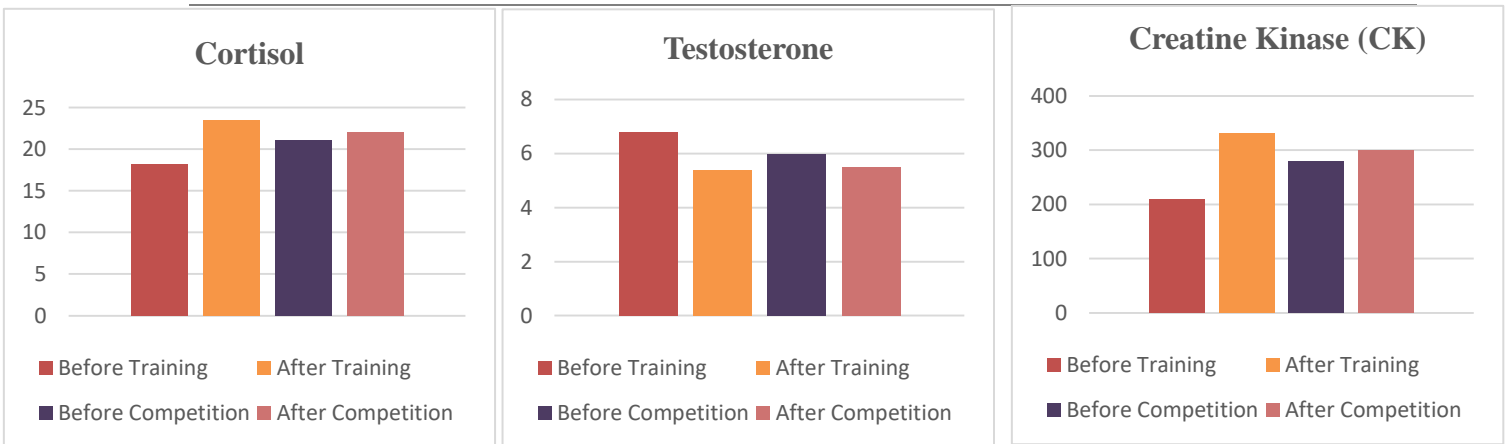
Data analysis will be conducted using SPSS (ver.26) software. Descriptive statistics will first be used to calculate the mean, standard deviation, and range for each biomarker at each time point. Paired t-tests will be performed to compare the levels of (cortisol, testosterone, and CK between) the four time points to assess how these biomarkers are affected by training and competition. A significance level of $p < 0.05$ will be considered statistically significant for all analyses.



Result

Table3: Descriptive Statistics and Statistical Comparison of Biomarkers at Different Phases of Training and Competition

Variable	Before Training		After Training		Before Competition		After Competition		t-value	p-value				
	Mean	SD	Mean	SD	Mean	SD	Mean	SD						
Cortisol	18.2	1.8	23.5	2.5	21.0	2.2	22.1	2.3	3.47	0.004				
Testosterone	6.8	0.5	5.4	0.4	6.0	0.4	5.5	0.3	2.16	0.032				
Creatine Kinase (CK)	210		10		330		15		280	12	300	20	5.12	0.0001



changes, which are induced by training and competition.

Cortisol Levels

Cortisol, the ant-stress hormone, also raised significantly after the training session, portraying excessive physiological stress under high impact workouts. Mean cortisol level increased from 18.2 $\mu\text{g/dL}$ before training to 23.5 $\mu\text{g/dL}$ after training ($t = 3.47$, $p = 0.004$). It is consistent with those of (Kreher and Schwartz 2012) who showed elevated cortisol levels following an acute bout of exercise over which the body is under stress. The slight variation in cortisol before (21.0 $\mu\text{g/dL}$) and

Figure1: Comparison of Biomarkers (Cortisol, Testosterone, and Creatine Kinase) Across Different Phases of Training and Competition

Discussion

This study gives a valuable indication on influence of different phases of training and competition on some of the key biomarkers: cortisol, testosterone and creatine kinase (CK) among professional wrestlers. These are biomarkers signaling stress, muscle recovery and muscle damage. Moreover, the relatively broad shifts found in these biomarkers underscore the substantial physiological



intensity resistance training and competitive events (indicative of muscle fiber damage). CK levels post exposition to competition were still significantly high, 300 U/L., correlating with the muscle stress as well as recovery requirement that occur during competition. The persisting increased CK levels after the competition reinforce those of (Dupuy et al. 2018), who reported that CK levels can remain high after intense physical activity, being related with delayed onset muscle soreness (DOMS).

Implications for Training and Recovery

the importance of athletic ability recovery after strenuous training and contests. Hormone and CK levels post- training are [much greater than] their pretreatment values, as indicating a lot of stress and muscle degradation in the wrestlers during exercising phase. Additionally, the reported rise in testosterone levels post exercise (supported by pharmacological data) combined with a decrease on running-induced serum testosterone and intratesticular testosterone indicates the existence of a temporary catabolic scenario that is disadvantageous to muscle regeneration and general performance. Such findings underscore the necessity of a well-designed training program and proper recovery to avoid overtraining while maximizing sport performance (Cadegiani et al., 2021).

The ability to track such biomarkers can help coaches and athletes understand the physiological stress from training loads, leading

after competition (22.1 $\mu\text{g/dL}$) suggests an adaptation to the reiterative activity of training and competing; this finding has been observed also by (Peake et al. 2017), when cortisol levels begin to plateau as the body adapts to the requirements of training.

Testosterone Levels

Testosterone a hormone necessary for muscle repair and growth, was significantly reduced during the training period. The average of testosterone level dropped from 6.8 ng/dL before exercise to 5.4 ng/dL after the exercise ($t = 2.16, p = 0.032$). This decrease is compatible with the results of Harman et al. (2001) who observed a decrease in testosterone after prolonged exercise. Testosterone levels just prior to competition (6.0 ng/dL) were still slightly above normal, and declined after the race (5.5 ng/dL). (Kraemer et al., 2005) observed that testosterone is not completely restored after intensive competitive schedules and maintain that the training and/or competition continues to place a heavy burden on the athlete's endocrine system.

Creatine Kinase (CK) Levels

CK, muscle damage biomarker, presented significant increase after training and competition periods. The mean CK rose from 210 U/L pre-training to 330 U/L post training ($t = 5.12, p = 0.0001$). This rise in CK activity correlates with the observations of (Simmons et al. 2021) who reported a comparable elevation in CK concentrations after high-



following the exercise, testosterone levels were dramatically reduced in what appears to be a partly catabolic state, which may be suboptimal for muscle recovery. On the other hand, testosterone levels partially recovered pre-competition but it got dropped again post-competition due to combined stress of training and competition. As expected, CK responses were strongly increased following exercise and both recruitment components showed substantial muscle damage during these phases. The increased CK levels following competition may further indicate the effect of a competitive environment on muscle stress. The results showed the importance of following these biomarkers in order to optimize training loads and recovery strategies. The results also indicate that the risk of overtraining could be increased if effective recovery strategies are not established, especially following periods of intense training and competition when cortisol and CK rise and testosterone falls.

Recommendations

Recovery between competition and hard training is important to allow high-performance levels without carrying over fatigues that might lead to overtraining. Extended monitoring of circulating biomarkers, such as cortisol, testosterone and creatine kinase (CK), promises to offer real-time information regarding the kinetics of physiological stress in an effort to guide training prescriptions accordingly. The use of periodized training will help limit every other dumping syndrome and thus help prevent overtraining. Active recovery (light exercise

to potential adaptations in physical preparation or recovery strategies.

This points towards the importance to explore large physiological changes in cortisol, testosterone and creatine kinase (CK) throughout various stages of a training and competition season. Elevated levels of these biomarkers are described as strong indicators of stress, recovery and muscle damage suggesting the importance of carefully controlled training loads and sound recovery practices for improving performance outcomes without risking overtraining. Future studies should explore recovery interventions with a more specific focus, to limit the negative consequences of excessive cortisol and CK levels, to allow prolonged development of athletes.

Conclusions

The present study demonstrates, with significant magnitude that professional wrestlers undergo important physiological modifications in the different phases of training and competition. Cortisol, testosterone and creatine kinase (CK) were measured as biomarkers of stress, muscle recovery and muscle damage. Training also caused key stress hormone levels such as cortisol, to be significantly higher after training than at rest, which is a high indicator of the stress created by hard physical exertion. This slight decrease in cortisol levels before and after the competition may be related to habituation from regular training and competition. Immediately



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and stretching) assists with muscle recovery and lowers blood CK levels. Similarly, implementing mental stress management strategies, such as mindfulness and meditation is valuable given their impact in managing cortisol levels, which vastly influence recovery.

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