Ivan Perasović[1]
Antonela Karmen Ivišić[1]
Dario Vrdoljak[1]*
Nikola Foretić[1,2]
Vladimir Pavlinović[1]
Mia Perić[1]
Zoran Nikolovski[1,2]

BIOMARKER DYNAMICS DURING OLYMPIC WEIGHTLIFTING COMPETITION; GENDER DIFFERENCES STUDY

DINAMIKA BIOMARKERJEV MED OLIMPIJSKIM DVIGOVANJEM UTEŽI; RAZISKAVA SPOLNIH RAZLIK

ABSTRACT

Sports competitions are defined as highly stressful environments, especially for the athletes. Apart from psychological, stressors perceived are connected with physical exertion and physiological response. Therefore, this study aimed to investigate physiological response following dynamics of Cortisol (C), alpha-amylase (AA) and testosterone (T) biomarkers during official olympic weightlifting (OW) competition. Additionally, the goal was to evaluate the difference between OW disciplines and the competition stress impact on athletes' nervous and endocrine systems. Lastly, the study aimed to determine the differences according to gender. The sample of participants in this study was 44 weightlifters (17 females). Their mean chronological age was 26.91 ± 7.57 years, body mass of 83.70 ± 23.01 kg, body height of 174.55 ± 11.92 cm, and training experience of 6.62 ± 6.95 years. In this study, saliva samples were collected during an official weightlifting competition and the AA activity and C and T concentrations were determined. The results showed that males have higher levels of T in pre-competition (males, 169.23 ± 52.37; females, 82.93 ± 23.46), post-snatch (males, 193.78 ± 56.77; females, 105.79 ± 36.18) and post-clean and jerk measurement (males, 177.63 ± 54.63; females, 107.00 ± 36.26). However, the percentage of changes shows that females have a higher increase of T through the competition (pre-competition to post-snatch, 27.56%; pre-competition to post-clean and jerk 29.02%) compared to males (pre-competition to post-snatch, 14.51%; pre-competition to post-clean and jerk 4.96%). Conclusively, Olympic weightlifting elicits both endocrine and nervous system activity through measured biomarkers. Furthermore, the snatch is observed as more stressful than clean and jerk, for both genders. Also, females have a greater percentage of increase in T values with lower absolute levels. Hence, knowing the impact of OW on athletes' stress different stress coping techniques should be applied in this sport.

Keywords: snatch, clean and jerk, testosterone, cortisol, alpha-amylase

[1]Faculty of Kinesiology, University of Split, 21000 Split, Croatia

[2]High Performance Sport Center, Croatian Olympic Committee, 10000 Zagreb, Croatia

IZVLEČEK

Športna tekmovanja so opredeljena kot visoko stresna okolja, še posebej za športnike. Poleg psihološkega so zaznani stresorji povezani s fizičnim naporom in fiziološkim odzivom. Zato je bil cilj te študije raziskati dinamiko biomarkerjev kortizola (C), alfa-amilaze (AA) in testosterona (T) med uradnim tekmovanjem v olimpijskem dvigovanju (OW) uteži. Poleg tega je bil cilj oceniti razlike med disciplinami olimpijskega dvigovanja uteži ter vpliv tekmovalnega stresa na živčni in endokrini sistem športnikov. Nazadnje je študija želela določiti razlike glede na spol. Vzorec udeležencev v tej študiji je bil 44 dvigovalcev uteži (17 žensk). Njihova povprečna kronološka starost je bila 26,91 ± 7,57 let, telesna masa 83,70 ± 23,01 kg, telesna višina 174,55 ± 11,92 cm in trenažna izkušenost 6,62 ± 6,95 let. V tej študiji so bile med uradnim tekmovanjem določene koncentracije C, AA in T. Rezultati so pokazali, da imajo moški višje ravni T v predtekmovalnem obdobju (moški, 169,23 ± 52,37; ženske, 82,93 ± 23,46), po nalogu potega (moški, 193,78 ± 56,77; ženske, 105,79 ± 36,18) in po nalogu izmeta (moški, 177,63 ± 54,63; ženske, 107,00 ± 36,26). Vendar pa odstotek sprememb kaže, da imajo ženske večje povečanje vrednosti T skozi tekmovanje (od predtekmovanja do po nalogu potega, 27,56 %; od predtekmovanja do po nalogu izmeta, 29,02 %) v primerjavi z moškimi (od predtekmovanja do po nalogu potega, 14,51 %; od predtekmovanja do po nalogu izmeta, 4,96 %). Sklepno, olimpijsko dvigovanje uteži povzroča aktivnost tako endokrinega kot živčnega sistema preko merjenih biomarkerjev. Poleg tega je bil nalog potega opažen kot bolj stresen kot nalog izmeta za oba spola. Prav tako imajo ženske večji odstotek povečanja vrednosti T z nižjimi absolutnimi vrednostmi. Zato ob poznavanju vpliva OW na stres športnikov je potrebno v tem športu uporabiti različne tehnike obvladovanja stresa.

Ključne besede: poteg, izmet, testosteron, kortizol, alfa-amilaza

Corresponding author:* Dario Vrdoljak

Faculty of Kinesiology, University of Split, 21000 Split, Croatia
E-mail: dariovrdoljak42@gmail.com
https://doi.org/10.52165/kinsi.31.1.52-68

INTRODUCTION

During sports competitions athletes are subject to high stress, triggering physiological and behavioural responses (Carrasco Páez & Martínez-Díaz, 2021). These responses can influence performance due to exertion-induced hormonal secretion and nervous system activity (Arai et al., 1989; Fletcher & Hanton, 2001; Furlan et al., 1993; Mellalieu & Juniper, 2006). Therefore, monitoring stress biomarkers, such as alpha-amylase (AA), cortisol (C) or testosterone (T), could lead to a more precise identification of factors influencing optimal athlete performance. Previously, it was shown that an anticipatory increase in C before sports competition is important for preparing athletes for psychological and physiological demands (Kjell et al., 2017). Furthermore, C was suggested to influence sports performance through its effect on cognitive processes, such as the activation and deactivation of the prefrontal cortex and amygdala (Bishop et al., 2004; Dedovic et al., 2009). Physical activity showed a different influence on other biomarkers. Whereas, T usually increases due to catabolic nature of physical activity (Kraemer & Rogol, 2008). Also, AA showed an increase due to nervous system's response to physical activity, which heightened sympathetic activity during high-intensity activities (Allgrove et al., 2008; Bishop et al., 2000)

C serves as an indicator of the activation of the hypothalamic-pituitary-adrenocortical axis, which plays a key role in the physiological response to stress (Stajer et al., 2020). It is commonly utilized as a biomarker to assess both physical and psychological stress in athletes (Moreira et al., 2012; Vitale et al., 2020). In support of this, Arruda and colleagues have conducted multiple studies on C responses in basketball, linking them to the psycho-physiological stress experienced by athletes, and have identified a correlation between C levels and players' anxiety (Arruda et al., 2014; Arruda et al., 2018).

T is an anabolic hormone that plays a crucial role in promoting the growth of skeletal muscle. Its effects are particularly significant during competitive events, where it appears to be vital for enhancing performance capabilities (Kraemer & Rogol, 2008). Research has indicated that T levels rise during and after competitions in sports such as basketball, Australian football, and soccer (Arruda et al., 2014; Arruda et al., 2018; Cormack et al., 2008; Fothergill et al., 2017). However, studies on American football have not found evidence of increased T levels as a result of competition (Hoffman et al., 1996).

AA is an enzyme that facilitates the breakdown of starch into smaller carbohydrate units, such as maltose (Granger et al., 2007). Exercise has been consistently shown to elevate both the activity and concentration of AA (Koibuchi & Suzuki, 2014), with the effect being particularly pronounced at exercise intensities exceeding 70% of VO2max (Koibuchi & Suzuki, 2014). Additionally, AA serves as a marker for sympathetic nervous system activity and is closely associated with increases in noradrenaline, thereby reflecting the state of arousal (Nater et al., 2007).

Accordingly, Olympic weightlifting (OW) is intertwined with physical, physiological and psychological stressors. OW consists of two primary lift disciplines: the snatch and the clean and jerk. These movements require a combination of power, speed, coordination, and flexibility, making technical execution just as critical as raw strength (Haff & Haff, 2019). Additionally, the intense neuromuscular demands of maximal-effort lifts cause substantial central nervous system (CNS) fatigue (Barnes et al., 2019). Following this physical and physiological stressors are quite obvious. On the other hand, psychological stress in OW is influenced by multiple factors. One of which is performance pressure, as athletes must manage expectations from coaches, teammates, and themselves, which can significantly elevate stress (Storey et al., 2016). Also, success in Olympic weightlifting is also determined by a lifter's ability to make strategic choices throughout the competition. This includes careful attempt selection, adjusting weights based on competitors' performances, and managing fatigue to optimize output across all six attempts. Lastly, athletes must also navigate the psychological pressures of competition, where factors such as crowd influence, time constraints, and expectations can impact performance (Mellalieu et al., 2009).

Following all of the abovementioned, the main aim of this study was to investigate the dynamics of C, AA and T biomarkers during official OW competition. Furthermore, to evaluate the difference between OW disciplines and the competition stress impact on athletes' nervous and endocrine systems. Lastly, the study aimed to determine the differences according to gender.

METHODS

Participants

The sample of participants included 44 weightlifters (17 female and 27 males) who regularly compete in national and international weightlifting competitions. Their mean chronological age was 26.91 ± 7.57 years, body mass of 83.70 ± 23.01 kg, body height of 174.55 ± 11.92 cm, and training experience of 6.62 ± 6.95 years. The participants were divided according to gender (see Fig 1.).

Table 1. Descriptive parameters of anthropometric and age data.

Variable	Males (N=27)		Females (N=17)	
	Mean	SD	Mean	SD
Age (years)	27.89	8.44	25.35	5.84
Body height (cm)	180.78	8.81	164.65	9.28
Body mass (kg)	95.73	19.87	64.60	12.24
Training experience (years)	8.37	8.05	3.84	3.34

Notes. SD=standard deviation.

All athletes who took part in this study volunteered and were informed about the purpose of the study. The participants were asked about the existence of an anxiety disorder or a tendency to panic anxiety, which was reported as non-existent. Experimental procedures were completed following the declaration of Helsinki, and they were approved by the Faculty of Kinesiology, University of Split ethics board (Ethics Board Approval No. 2181-205-02-5-23-027).

Procedure

Salivary biomarkers were measured at three-time points: before the start of the competition (8:00 – 10:00 AM), after the third snatch attempt, and after the third clean and jerk attempt (see Figure 1). Due to the competition's nature, the time frame between the second and third measurements varied by approximately 1 hour. Precisely, after official weighting, competitors started their usual warm-up. They reported their starting weights for both snatch and clean and jerk discipline. Following that, they competed in the snatch discipline with three lifts per competitor. If they managed to clear the first weight they proceeded to higher weights. After all participants finished with snatch they had 20 minutes of break between disciplines. Lastly, the clean and jerk was done in the same manner as the snatch with three attempts per weight. Apart from salivary biomarkers, the study variables included anthropometric measurements. Body mass (measured to the nearest 0.1 kg) and body height (measured in cm) were assessed using a stadiometer (Seca 213, Hamburg, Germany) and a digital scale (Seca 769, Hamburg, Germany).

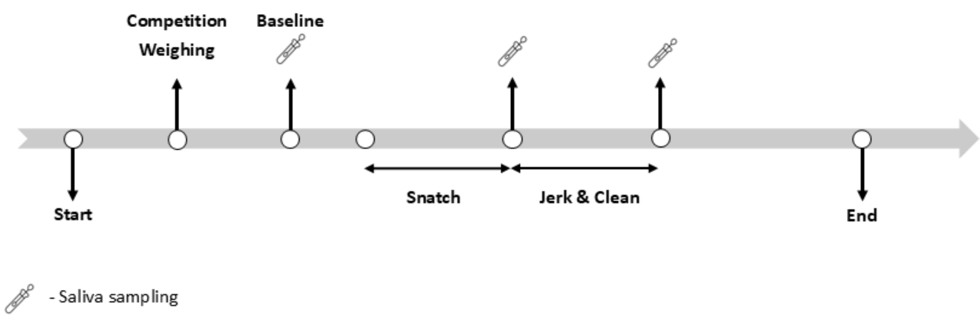

Figure 1. Sample collection timeline before the competition and directly snatch and clean and jerk.

Sampling and Handling

Saliva samples were collected using SalivaBio Oral Swabs (Salimetrics LLC, State College, PA, USA), placed under the tongue for 2 minutes, and immediately refrigerated. Participants were instructed to fast overnight and avoid major meals 60 minutes before sample collection. In order to standardize procedures, participants rinsed their mouths with water 10 minutes before sampling and refrained from consuming fluids during this period. After collection, the swabs were placed into a storage tube and refrigerated immediately. Within 2 hours following sampling, samples were frozen at below –20 °C until centrifugation. On the day of analysis, the samples were thawed completely and centrifuged at 1500 x g (3000 rpm) for 15 minutes. After centrifugation, assays were performed. Saliva C and T were analyzed with a commercially available enzyme-linked immunosorbent assay (ELISA) purchased from Salimetrics LLC (State College, PA, USA), on a microplate reader (Infinite 200PRO, Tecan, Mannendorf, Switzerland). Standard curves were constructed according to the manufacturer's instructions and commercially available standards, and quality control samples were used for all assays (Salimetrics LLC). The assay sensitivity for salivary T was 1 pg/ml, while the average intra-assay coefficient of variation (CV) was 4.6%, with a CV of duplicate analysis of 3.8%. The assay sensitivity for salivary C was 0.007 µg/dl, with an average intra-assay CV of 4.5%. All samples were analyzed in the same batch to avoid intraassay variability. Samples for AA were analyzed using a kinetic enzyme assay kit from the same supplier (Salimetrics LLC, State College, PA, USA). The average intra-assay CV was 5.5%. Values are expressed as the AA concentration (U/ml). Salivary hormone concentrations were corrected for the salivary flow rate. All data were log-transformed to reduce the non-uniformity of error, and normality was tested using the Kolmogorov–Smirnov test procedure. Homoscedasticity was checked by the Levene test.

Statistical analysis

The statistical analyses included the calculation of descriptive statistics with means and standard deviations. The differences among measurements of salivary biomarkers were calculated with a dependent t-test to determine differences between OW disciplines. Following that, the independent sample t-test was used to determine possible gender differences. The t-test's p-values were checked by the magnitude-based Cohen's D effect size (ES) statistic. Modified qualitative descriptors of ES were (trivial ES: < 0.2; small ES: 0.21–0.60; moderate ES:0.61–1.20; large ES: 1.21–1.99; and very large ES: > 2.0 (Cohen, 1988).

The software Statistica ver. 13.0 (Dell Inc, USA) was used for all analyses, and a p-level of 95% ($p < 0.05$) was applied.

RESULTS

The analysis of Table 2 shows differences between males and females in all three measured biomarkers (AA, C, and T) on 3 selected time points (pre-competition, post-snatch and post-jerk & clean). The differences are observed only in T. Precisely, males showed higher levels of T in in pre-competition (males, 169.23 ± 52.37; females, 82.93 ± 23.46), post-snatch (males, 193.78 ± 56.77; females, 105.79 ± 36.18) and post-clean and jerk measurement (males, 177.63 ± 54.63; females, 107.00 ± 36.26). However, the percentage of changes shows that females have higher increase of T through the competition (pre-competition to post-snatch, 27.56%; pre-competition to post-clean and jerk 29.02%) compared to males (pre-competition to post-snatch, 14.51%; pre-competition to post-clean and jerk 4.96%).

Table 2. Gender differences in AA, C and T in all measured time points.

Variable	Males (N=27)		Females (N=17)		t	p	ES	95% CI
	Mean	SD	Mean	SD				
AA_pre (U/ml)	155.87	120.54	122.00	124.11	0.88	0.38	0.28	-0.36 – 0.92
AA_post snatch (U/ml)	286.59	285.64	220.64	187.29	0.84	0.40	0.26	-0.37 – 0.89
AA_post jerk & clean (U/ml)	288.89	209.49	227.46	152.18	1.05	0.30	0.32	-0.30 – 0.95
C_pre (µg/dL)	0.34	0.20	0.27	0.07	1.48	0.15	0.46	-0.18 – 1.11
C_post snatch (µg/dL)	0.62	0.20	0.54	0.20	1.24	0.22	0.38	-0.25 – 1.01
C_post clean and jerk(µg/dL)	0.42	0.21	0.50	0.19	-1.38	0.18	0.43	-0.21 – 1.06
T_pre (pg/ml)	169.23	52.37	82.93	23.46	6.36	0.00*	2.00[¥]	1.22 – 2.77
T_post snatch (pg/ml)	193.78	56.77	105.79	36.18	5.41	0.00*	1.74[¥]	0.99 – 2.50
T_post clean and jerk (pg/ml)	177.63	54.63	107.00	36.26	4.69	0.00*	1.46[¥]	0.76 – 2.17

Notes. AA=Alpha-amylase; C=Cortisol; T=Testosterone; SD=standard deviation; t=test value; p=level of significance set at p<0.05; ES=Effect size; CI=confidence intervals; *=significance; ¥=high ES.

The analysis of results in Figure 2 shows differences between measurements of AA, C and T for all participants, and males and females separately. In AA moderate ES is observed between pre-competition and post-snatch in all participants (0.66) and females (0.62). Similar ES is calculated in pre-competition and post-clean and jerk (all, 0.77; females, 0.76). In male group the moderate ES is found between pre-competition and post-snatch measurement (0.77). Furthermore, observation of C dynamics showed high ES between pre-competition and post-snatch (all, 1.50; males, 1.38; females, 1.80), and pre-competition and post-clean and jerk only in females (1.62). Also, moderate ES is seen between pre-competition and post-clean and jerk (0.74) and between post-snatch to post-clean and jerk (0.67), in all participants. Male group had moderate ES between post-snatch to post-clean and jerk (0.97). In T only females had moderate ES in pre-competition to post-clean and jerk (0.76) and between pre-competition to post-clean and jerk (0.79).

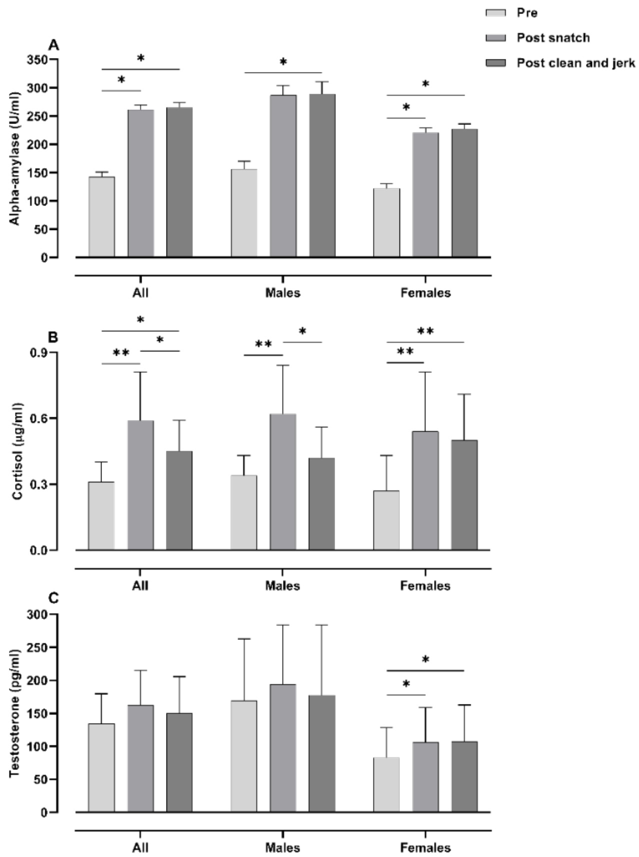

Figure 2. Differences among different measurement points in all three biomarkers, for alpha-amylase (A), cortisol (B) and testosterone (C). With moderate effect size marked as *; and high effect size marked as **.

The analysis of results in Figure 3 shows percentage changes for all participants, males and females in all three measured biomarkers. The results indicate a similar percentage of increase in AA for pre to post-snatch (all, 136.75 %; males, 125.39 %; females, 153.45 %) and in pre to post-clean and jerk (all, 136.99 %; males, 122.21 %; females, 158.71 %). Oppositely, C for pre to post-snatch (all, 125.20 %; males, 131.10 %; females, 116.52 %) shows a higher increase than in pre to post-clean and jerk (all, 69.03 %; males, 49.55 %; females, 97.67 %). Lastly, the a similar trend between pre to post-snatch (15.39 %) and pre to post-clean and jerk (16.75 %) in all participants. However, in a group of male participants perceived a higher increase in pre to post-snatch (19.25 %) than pre to post-clean and jerk (7.36 %). Whereas, females have the opposite trend showing a higher increase in pre to post-clean and jerk (30.58 %) than in pre to post-snatch (9.71 %).

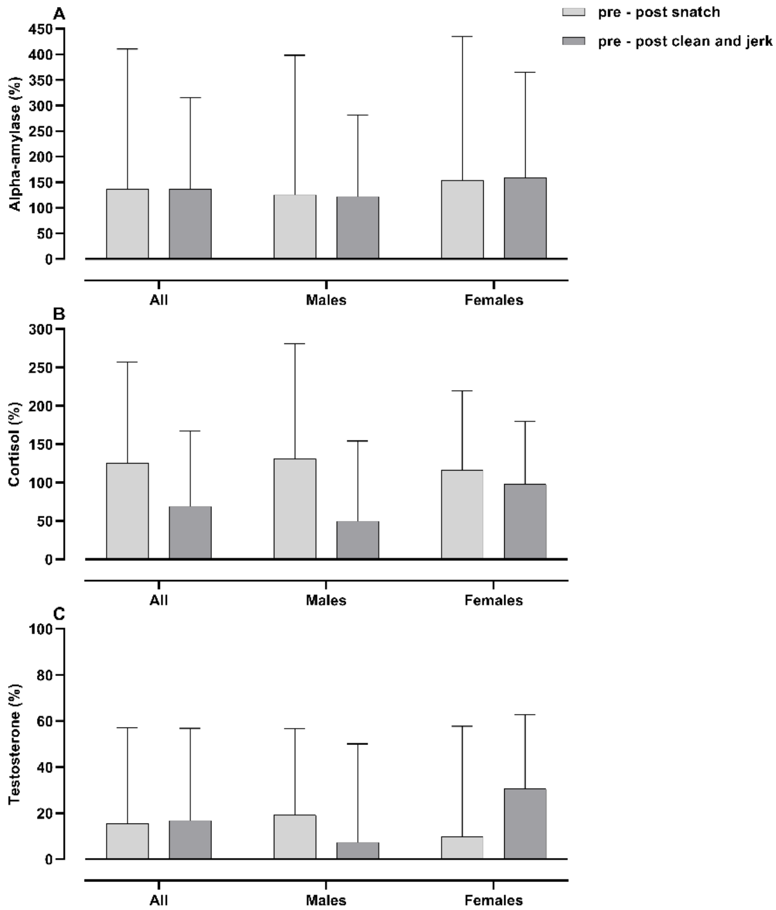

Figure 3. Percentage change among different measurement points in all three biomarkers, for alpha-amylase (A), cortisol (B) and testosterone (C).

DISCUSSION

Following the aims of the study, a few credible main findings could be reported: (i) weightlifting competition elicits an increase in AA and C, whereas an increase in T is seen only in female athletes.; (ii) The males and females differ only in T dynamics; (iii) baseline values of T are significantly lower, with higher percentage of increase during competition in females.

Gender differences between female and male weightlifters

Given that there are no notable differences in the baseline values of C and AA, and considering they exhibit the same trends and comparable absolute values of these biomarkers throughout the competition, it can be stated that female and male weightlifters have similar stress response during OW competition. These similarities are further supported by the absence of significant differences found through t-tests and effect size calculations.

Examining prior research on gender differences in stress reactions related to sports competition reveals a lack of consistency. Some studies reported higher C and AA responses in females and some in male athletes (Aubets & Segura, 1995; Filaire et al., 2009; L. Peñailillo, 2018; Mroczkowska et al., 2020). Conversely, research on younger athlete populations has found similar competition stress responses in both genders. This has been observed in studies involving adolescent Olympic weightlifters, elite junior golfers, university students, and collegiate wrestlers. (Crewther et al., 2016; Kim et al., 2010; Kobayashi et al., 2024; Trochimiak et al., 2015). Mentioned studies focus on individual sports and physical activities that are relatively short in duration and high in intensity, resembling OW competition.

Given that most researchers recognize the type of physical activity as a critical factor influencing the stability of stress responses, it is likely that this aspect played a significant role in our study as well (Aubets & Segura, 1995; Chiodo et al., 2011; Crewther et al., 2016; Edmonds et al., 2016). OW stands out as a sport where volume and intensity remain relatively stable in competition (i.e., six attempts at maximal or near-maximal loads), unlike many other sports where these variables fluctuate unpredictably. In summary, the more predictable the activity, the more anticipated stress response, making it less affected by the athlete's individual reactions to competition. This may help explain the comparable C and AA responses observed in both male and female weightlifters in our research.

Analysis of gender differences when it comes to T yields seemingly different insights. Although significant differences were found at all measurement points (before, during and after the competition), the change trends are very similar in both female and male lifters. Hence, differences are primarily influenced by the initial T level, which is naturally significantly higher in male lifters. Men generally have higher baseline T levels than women. This is due to the naturally higher T levels in men, which are essential for muscle growth and strength development. Although women have lower baseline T levels, they remain sufficient to sustain athletic performance and muscle maintenance (Kraemer & Ratamess, 2005; Le Panse et al., 2012). Ignoring baseline differences, T levels increased similarly after the snatch during competition in male and female lifters, respectively. However, following the clean and jerk, T levels continued to rise in female while slightly declining in male lifters. This indicates that OW competition requires a higher level of T secretion in female weightlifters compared to other physical activities, such as soccer or resistance training reported in previous studies (Edwards et al., 2006; Vingren et al., 2009). Regarding all above-mentioned in terms of gender differences, we can assume that OW competition elicits equal T, C, and AA responses in male and female Olympic weightlifters.

Biomarkers dynamics during Weightlifting competition

When observing the dynamics of measured biomarkers (AA, C, and T) it can be noted that they have similar trends concerning gender. However, each biomarker follows its individual fluctuation pattern. Precisely, AA results indicate a high increase after snatch and it stays high for the rest of the competition. Such an increase indicates that snatch is highly stressful for the nervous system (Collet et al., 2006; Passelergue et al., 1995). The highly impactful influence of snatch on AA can be observed through the nature of this OW discipline. Previous studies demonstrate that snatch relies more on the explosiveness of the athlete, more than clean and jerk, which is dominantly influenced with athelets maximal strength (Joffe et al., 2023; Sandau & Kipp, 2025). On the other hand, the high levels of AA after clean and jerk should be observed with caution, since the measurement after snatch and before clean and jerk is lacking. Nevertheless, both disciplines are eliciting nervous system activation. Additionally, observation of the diurnal rhythm of biomarkers showed that in a normal daily routine AA is in its lower domain during morning hours when OW competition occurs (Nater et al., 2007). Such rhythm shows that the increase of AA is indeed due to the high intensity of OW. Similarly, previous studies showed that AA increases due to both psychological (Foretić et al., 2022; Nikolovski et al., 2024) and physiological stress (Foretic et al., 2020; Foretić et al., 2023). Furthermore, OW competitions lack certain „surprise factors“. Therefore, the physical component of the sport itself is influencing nervous system stress. Hence, it can be concluded that OW competition is highly stressful for these athletes in both disciplines, regarding AA increase.

A similar trend is visible in T during throughout the competition. Precisely, T increases and remains high until the end of the competition. Previous literature (Foretic et al., 2020; Foretić et al., 2023), reported that depending the nature of sport, especially in the contact domain, T increases significantly. Barnes et al. (2012) showed that rugby players experience high T rise during both match and training sessions. Therefore, the increase in T is connected with internal and external workload (Gaviglio et al., 2015). Observing OW competition as such, it can be noted that both males and females experience an increase in T. Following the abovementioned, this increase could be seen as athletes' response to OW as a physically demanding sport. Also, high elicitation of motivation and aggression is a valuable factor for performance in OW. According to Gleason et al. (2009), T acts on specific substrates in the brain to increase aggression and motivation during competition (Öktem & Kul, 2020). On the other hand, the increase of T is visible after the snatch, the values decrease after clean and jerk indicating the different responses of athletes regarding discipline. Previously the connection between AA and T has been defined (Kivlighan & Granger, 2006). These studies explained that an increase in AA will elicit the secretion of T (Granger et al., 2007; Kivlighan & Granger, 2006). Similar to AA trends, the nervous system activity in snatch is more pronounced than in clean and jerk which explains the mentioned increase-decrease trend of T.

This trend of increase and decrease regarding discipline is even more seen in C secretion. The secretion of this biomarker is highly connected to endocrine system activity (Nater et al., 2006; Nater et al., 2005). With that, it is released at a slower level than AA or T (Nater et al., 2005). Since weightlifters have 3 attempts per discipline which last up to 1 min per attempt, the time to elicit C is not sufficient for higher increases. Previous studies showed that playing time has of high influence on C levels (Foretic et al., 2020; Foretić et al., 2023; Nikolovski et al., 2023). More precisely, athletes who have a longer time in physical activity showed higher levels of C. Additionally, the physical component of OW is high and physical stresses are pronounced (Storey et al., 2016). Once again, this situation could explain the trend of C. Also, the diurnal rhythm of C shows that in the hours of the competition, it is in an increasing trend (Nater et al., 2007), which could influence the level of secretion. Afterwards, the normal levels at which C stabilizes are reached. This normal daily rhythm could also explain the increase of C after snatch and decrease post clean and jerk.

Strengths and limitations

The main limitation of this study is the lack of baseline biomarker values on non-competitive days, which sets precautions for the interpretation of the results. Also, the circadian rhythm of the measured biomarkers can be seen as a limitation. This rhythm alters the usual daily fluctuations of the biomarkers, potentially affecting measurements and result analysis. Additionally, the half-life (duration of biomarker presence in the human body) of biomarkers could be seen as a limitation. Since OW competition is short in duration, hence the secretion of biomarkers, which vary in time, could be diminished or postponed (especially in C values). Additionally, since this study included female athletes, the menstrual cycle's influence on measured biomarkers should have been explored. However, this study is the first to examine a sample of Olympic weightlifters during official national competition. Also, a relatively high sample of participants is a strength of the study. With that being said, the ecological validity of this study is the main strength of this study.

CONCLUSION

Research results demonstrate how an OW competition influences the secretion of salivary biomarkers (AA, C, and T). The study aimed to define the dynamics of biomarkers under the influence of OW competition. Furthermore, to determine the gender differences for measured biomarkers. Participants in this study perceived an increasing trend of AA after snatch discipline, and after clean and jerk the values remained the same. T showed a similar trend for both disciplines, whereas C showed an increase and then significantly decreased post-clean and jerk. The dynamics do not differ significantly between genders. However, when genders are compared, the only significance was observed in females for T values. Also, significantly higher baseline T levels in male weightlifters created differences in all measuring points nevertheless of same trends in both genders.

To conclude, it can be stated that OW disciplines differently influence measured biomarkers. Precisely, snatch showed to be a more pronounced stressor that influenced both the endocrine and nervous systems. Additionally, even though the obtained gender differences showed that females have lower absolute values of T, the differences are lacking when the relative increase is observed. Hence, knowing the impact of OW on athletes, different stress coping techniques could be used (slow-paced breathing, diaphragmatic breathing) to manipulate stress levels and improve their performance. This could help in functional stress level manipulation and improvement of competitive performance. Also, the practical application of this study is observed in the possibility to influence stress in OW athletes and to establish the stress which occurs differently in regard to disciplines and gender.

Declaration of Conflicting Interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

REFERENCES

Allgrove, J. E., Gomes, E., Hough, J., & Gleeson, M. (2008). Effects of exercise intensity on salivary antimicrobial proteins and markers of stress in active men. *Journal of Sports Sciences*, *26*(6), 653-661. https://doi.org/10.1080/02640410701716790

Arai, Y., Saul, J. P., Albrecht, P., Hartley, L. H., Lilly, L. S., Cohen, R. J., & Colucci, W. S. (1989). Modulation of cardiac autonomic activity during and immediately after exercise. *American Journal of Physiology-Heart and Circulatory Physiology*, *256*(1), H132-H141. https://doi.org/10.1152/ajpheart.1989.256.1.H132

Arruda, A. F. S., Aoki, M. S., Freitas, C. G., Drago, G., Oliveira, R., Crewther, B. T., & Moreira, A. (2014). Influence of competition playing venue on the hormonal responses, state anxiety and perception of effort in elite basketball athletes. *Physiology and Behavior*, *130*, 1-5. https://doi.org/https://doi.org/10.1016/j.physbeh.2014.03.007

Arruda, A. F. S. d., Aoki, M. S., Paludo, A. C., Drago, G., & Moreira, A. (2018). Competition stage influences perceived performance but does not affect rating of perceived exertion and salivary neuro-endocrine-immune markers in elite young basketball players. *Physiology and Behavior*, *188*, 151-156. https://doi.org/https://doi.org/10.1016/j.physbeh.2018.02.009

Aubets, J., & Segura, J. (1995). Salivary cortisol as a marker of competition related stress. *Science & Sports*, *10*(3), 149-154.

Barnes, M. J., Miller, A., Reeve, D., & Stewart, R. J. C. (2019). Acute Neuromuscular and Endocrine Responses to Two Different Compound Exercises: Squat vs. Deadlift. *The Journal of Strength & Conditioning Research*, *33*(9). https://journals.lww.com/nsca-jscr/fulltext/2019/09000/acute_neuromuscular_and_endocrine_responses_to_two.10.aspx

Barnes, M. J., Mundel, T., & Stannard, S. R. (2012). The effects of acute alcohol consumption on recovery from a simulated rugby match. *Journal of Sports Sciences*, *30*(3), 295-304. https://doi.org/10.1080/02640414.2011.637949

Bishop, N. C., Blannin, A. K., Armstrong, E., Rickman, M., & Gleeson, M. (2000). Carbohydrate and fluid intake affect the saliva flow rate and IgA response to cycling. *Medicine and science in sports and exercise*, *32*(12), 2046-2051. https://doi.org/10.1097/00005768-200012000-00013

Bishop, S., Duncan, J., Brett, M., & Lawrence, A. D. (2004). Prefrontal cortical function and anxiety: controlling attention to threat-related stimuli. *Nature Neuroscience*, *7*(2), 184-188. https://doi.org/10.1038/nn1173

Carrasco Páez, L., & Martínez-Díaz, I. C. (2021). Training vs. Competition in Sport: State Anxiety and Response of Stress Hormones in Young Swimmers. *Journal of Human Kinetics*, *80*(1), 103-112. https://doi.org/doi:10.2478/hukin-2021-0087

Chiodo, S., Tessitore, A., Cortis, C., Cibelli, G., Lupo, C., Ammendolia, A., De Rosas, M., & Capranica, L. (2011). Stress-related hormonal and psychological changes to official youth Taekwondo competitions. *Scandinavian journal of medicine & science in sports*, *21*(1), 111-119.

Cohen, J. (1988). Statistical power analysis for the behavioural sciences. Hillsdale, New Jersey: L. *Lea*, *56*, 102.

Collet, C., Guillot, A., Bolliet, O., & Dittmar, A. (2006). Autonomic-Nervous-System Activity During the Preparation Phase for the Snatch in Olympic Weight Lifting. *International Journal of Sports Physiology and Performance*, *1*(4), 375-387. https://doi.org/10.1123/ijspp.1.4.375

Cormack, S. J., Newton, R. U., & McGuigan, M. R. (2008). Neuromuscular and endocrine responses of elite players to an Australian rules football match. *International Journal of Sports Physiology and Performance*, *3*(3), 359-374. https://doi.org/10.1123/ijspp.3.3.359

Crewther, B., Obminski, Z., & Cook, C. (2016). The effect of steroid hormones on the physical performance of boys and girls during an Olympic weightlifting competition. *Pediatric exercise science*, *28*(4), 580-587.

Dedovic, K., Duchesne, A., Andrews, J., Engert, V., & Pruessner, J. C. (2009). The brain and the stress axis: The neural correlates of cortisol regulation in response to stress. *NeuroImage*, *47*(3), 864-871. https://doi.org/https://doi.org/10.1016/j.neuroimage.2009.05.074

Edmonds, R., Leicht, A., Burkett, B., & McKean, M. (2016). Cardiac autonomic and salivary responses to a repeated training bout in elite swimmers. *Sports*, *4*(1), 13.

Edwards, D. A., Wetzel, K., & Wyner, D. R. (2006). Intercollegiate soccer: Saliva cortisol and testosterone are elevated during competition, and testosterone is related to status and social connectedness with teammates. *Physiology & behavior*, *87*(1), 135-143.

Filaire, E., Alix, D., Ferrand, C., & Verger, M. (2009). Psychophysiological stress in tennis players during the first single match of a tournament. *Psychoneuroendocrinology*, *34*(1), 150-157.

Fletcher, D., & Hanton, S. (2001). The relationship between psychological skills usage and competitive anxiety responses. *Psychology of Sport and Exercise*, *2*(2), 89-101. https://doi.org/https://doi.org/10.1016/S1469-0292(00)00014-5

Foretic, N., Nikolovski, Z., Peric, I., & Sekulic, D. (2020). Testosterone, cortisol and alpha-amylase levels during a handball match; analysis of dynamics and associations. *Res Sports Med*, *28*(3), 360-370. https://doi.org/10.1080/15438627.2020.1759069

Foretić, N., Nikolovski, Z., Marić, D., Gabrilo, G., Sekulić, D., Jaksić, D., & Drid, P. (2022). Stress Levels in Handball Coaching–Case Study: Preliminary Analysis of the Differences between Training and Match. *International Journal of Environmental Research and Public Health*, *19*(16), 10251.

Foretić, N., Nikolovski, Z., Marić, D., Perić, R., & Sekulić, D. (2023). Analysis of the associations between salivary cortisol-, alpha-amylase-, and testosterone-responsiveness with the physical contact nature of team handball: a preliminary analysis. *J Sports Med Phys Fitness*, *63*(1), 170-177. https://doi.org/10.23736/s0022-4707.22.13963-0

Fothergill, M., Wolfson, S., & Neave, N. (2017). Testosterone and cortisol responses in male soccer players: The effect of home and away venues. *Physiology and Behavior*, *177*, 215-220. https://doi.org/https://doi.org/10.1016/j.physbeh.2017.04.021

Furlan, R., Piazza, S., Dell'Orto, S., Gentile, E., Cerutti, S., Pagani, M., & Malliani, A. (1993). Early and late effects of exercise and athletic training on neural mechanisms controlling heart rate. *Cardiovascular Research*, *27*(3), 482-488. https://doi.org/10.1093/cvr/27.3.482

Gaviglio, C. M., Osborne, M., Kelly, V. G., Kilduff, L. P., & Cook, C. J. (2015). Salivary testosterone and cortisol responses to four different rugby training exercise protocols. *European Journal of Sport Science*, *15*(6), 497-504. https://doi.org/https://doi.org/10.1080/17461391.2015.1017012

Gleason, E. D., Fuxjager, M. J., Oyegbile, T. O., & Marler, C. A. (2009). Testosterone release and social context: When it occurs and why. *Frontiers in Neuroendocrinology*, *30*(4), 460-469. https://doi.org/https://doi.org/10.1016/j.yfrne.2009.04.009

Granger, D. A., Kivlighan, K. T., El-Sheikh, M., Gordis, E. B., & Stroud, L. R. (2007). Salivary α-Amylase in Biobehavioral Research. *Annals of the New York Academy of Sciences*, *1098*(1), 122-144. https://doi.org/https://doi.org/10.1196/annals.1384.008

Haff, G. G., & Haff, E. E. (2019). Weightlifting for young athletes. In *Strength and Conditioning for Young Athletes* (pp. 155-187). Routledge.

Hoffman, J. R., Tenenbaum, G., Maresh, C. M., & Kraemer, W. J. (1996). Relationship between athletic performance tests and playing time in elite college basketball players. *The Journal of Strength & Conditioning Research*, *10*(2), 67-71.

Joffe, S. A., Price, P., Chavda, S., Shaw, J., & Tallent, J. (2023). The Relationship of Lower-Body, Multijoint, Isometric and Dynamic Neuromuscular Assessment Variables With Snatch, and Clean and Jerk Performance in Competitive Weightlifters: A Meta-Analysis. *Strength & Conditioning Journal*, *45*(4). https://journals.lww.com/nsca-scj/fulltext/2023/08000/the_relationship_of_lower_body,_multijoint,.3.aspx

Kim, K.-J., Park, S., Kim, K.-H., Jun, T.-W., Park, D.-H., & Kim, K.-B. (2010). Salivary cortisol and immunoglobulin A responses during golf competition vs. practice in elite male and female junior golfers. *The Journal of Strength & Conditioning Research*, *24*(3), 852-858.

Kivlighan, K. T., & Granger, D. A. (2006). Salivary α-amylase response to competition: Relation to gender, previous experience, and attitudes. *Psychoneuroendocrinology*, *31*(6), 703-714. https://doi.org/https://doi.org/10.1016/j.psyneuen.2006.01.007

Kjell, N. v. P., Matthew, A. T., Charlotte, M. N., & Matt, B. (2017). The anticipatory stress response to sport competition; a systematic review with meta-analysis of cortisol reactivity. *BMJ Open Sport & Exercise Medicine*, *3*(1), e000261. https://doi.org/10.1136/bmjsem-2017-000261

Kobayashi, Y., Koibuchi, E., Sakuraba, K., & Suzuki, Y. (2024). Influence of AMY1 gene copy number on salivary amylase activity changes induced by exercise in young adults. *Physiological Reports*, *12*(20), e70099.

Koibuchi, E., & Suzuki, Y. (2014). Exercise upregulates salivary amylase in humans (Review). *Exp Ther Med*, *7*(4), 773-777. https://doi.org/10.3892/etm.2014.1497

Kraemer, W. J., & Ratamess, N. A. (2005). Hormonal responses and adaptations to resistance exercise and training. *Sports medicine*, *35*, 339-361.

Kraemer, W. J., & Rogol, A. D. (2008). *The endocrine system in sports and exercise*. John Wiley & Sons.

L. Peñailillo, F. A. E., Esteban R Jury, M. Castro-Sepulveda, L. Deldicque, H. Zbinden-Foncea. (2018). Differences in salivary hormones and perception of exertion in elite women and men volleyball players during tournament. *The Journal of sports medicine and physical fitness*, *58*(11), 6. https://doi.org/10.23736/S0022-4707.17.07681-2

Le Panse, B., Labsy, Z., Baillot, A., Vibarel-Rebot, N., Parage, G., Albrings, D., Lasne, F., & Collomp, K. (2012). Changes in steroid hormones during an international powerlifting competition. *Steroids*, *77*(13), 1339-1344.

Mellalieu, S. D., & Juniper, S. W. (2006). A Qualitative Investigation into Experiences of the Role Episode in Soccer. *The Sport Psychologist*, *20*(4), 399-418. https://doi.org/10.1123/tsp.20.4.399

Mellalieu, S. D., Richard, N., Sheldon, H., & and Fletcher, D. (2009). Competition stress in sport performers: Stressors experienced in the competition environment. *Journal of Sports Sciences*, *27*(7), 729-744. https://doi.org/10.1080/02640410902889834

Moreira, A., McGuigan, M. R., Arruda, A. F. S., Freitas, C. G., & Aoki, M. S. (2012). Monitoring Internal Load Parameters During Simulated and Official Basketball Matches. *The Journal of Strength & Conditioning Research*, *26*(3). https://journals.lww.com/nsca-jscr/Fulltext/2012/03000/Monitoring_Internal_Load_Parameters_During.37.aspx

Mroczkowska, H., Supiński, J., Litwiniuk, A., & Obmiński, Z. (2020). Biomedical indicators of pre-competitive stress in female and male judo players. SOCIETY. INTEGRATION. EDUCATION. Proceedings of the International Scientific Conference,

Nater, U. M., La Marca, R., Florin, L., Moses, A., Langhans, W., Koller, M. M., & Ehlert, U. (2006). Stress-induced changes in human salivary alpha-amylase activity—associations with adrenergic activity. *Psychoneuroendocrinology*, *31*(1), 49-58. https://doi.org/https://doi.org/10.1016/j.psyneuen.2005.05.010

Nater, U. M., Rohleder, N., Gaab, J., Berger, S., Jud, A., Kirschbaum, C., & Ehlert, U. (2005). Human salivary alpha-amylase reactivity in a psychosocial stress paradigm. *International Journal of Psychophysiology*, *55*(3), 333-342. https://doi.org/https://doi.org/10.1016/j.ijpsycho.2004.09.009

Nater, U. M., Rohleder, N., Schlotz, W., Ehlert, U., & Kirschbaum, C. (2007). Determinants of the diurnal course of salivary alpha-amylase. *Psychoneuroendocrinology*, *32*(4), 392-401. https://doi.org/https://doi.org/10.1016/j.psyneuen.2007.02.007

Nikolovski, Z., Foretić, N., Vrdoljak, D., Marić, D., & Perić, M. (2023). Comparison between Match and Training Session on Biomarker Responses in Handball Players. *Sports*, *11*(4), 83.

Nikolovski, Z., Vrdoljak, D., Foretić, N., Perić, M., Pavlinović, V., Perić, R., & Karanović, V. (2024). Levels of Stress in Volleyball Referees During Official Matches-The Influence of the Referee Role and Level of Competition. *Sports (Basel)*, *12*(12). https://doi.org/10.3390/sports12120319

Öktem, T., & Kul, M. (2020). Examining the Relationship between Self-Efficacy and Aggression Levels of Students Interested in Weightlifting Sports. *African Educational Research Journal*, *8*(4), 681-688.

Passelergue, P., Robert, A., & Lac, G. (1995). Salivary Cortisol and Testosterone Variations during an Official and a Simulated Weight-Lifting Competition. *Int J Sports Med*, *16*(05), 298-303. https://doi.org/10.1055/s-2007-973009

Sandau, I., & Kipp, K. (2025). Prediction of Snatch and Clean and Jerk Performance From Physical Performance Measures in Elite Male Weightlifters. *Journal of Strength and Conditioning Research*, *39*(1), 33-40. https://doi.org/10.1519/jsc.0000000000004945

Stajer, V., Vranes, M., & Ostojic, S. M. (2020). Correlation between biomarkers of creatine metabolism and serum indicators of peripheral muscle fatigue during exhaustive exercise in active men. *Research in Sports Medicine*, *28*(1), 147-154. https://doi.org/10.1080/15438627.2018.1502185

Storey, A. G., Birch, N. P., Fan, V., & Smith, H. K. (2016). Stress responses to short-term intensified and reduced training in competitive weightlifters. *Scandinavian Journal of Medicine and Science in Sports*, *26*(1), 29-40. https://doi.org/https://doi.org/10.1111/sms.12400

Trochimiak, T., Hübner-Wozniak, E., & Tomaszewski, P. (2015). The resting salivary antimicrobial proteins and cortisol concentration in wrestlers during 12-week training. *Biomedical Human Kinetics*, *7*(1).

Vingren, J. L., Kraemer, W. J., Hatfield, D. L., Volek, J. S., Ratamess, N. A., Anderson, J. M., Häkkinen, K., Ahtiainen, J., Fragala, M. S., & Thomas, G. A. (2009). Effect of resistance exercise on muscle steroid receptor protein content in strength-trained men and women. *Steroids*, *74*(13-14), 1033-1039.

Vitale, J. A., Povìa, V., Belli, E., Lombardi, G., Banfi, G., & La Torre, A. (2020). Are two different speed endurance training protocols able to affect the concentration of serum cortisol in response to a shuttle run test in soccer players? *Research in Sports Medicine*, *28*(2), 293-301. https://doi.org/10.1080/15438627.2019.1635131