

Cortisol awakening response is associated with fatigue following a single bench press exercise

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Abstract

Background: The cortisol awakening response (CAR) may reflect the physical adaptation to exercise and training loads, and recently a residual effect of a single high-intensity exercise has been reported. However, few studies have examined the effects of resistance exercise. The purpose of this study was to investigate the effect of a single high-intensity resistance exercise on the CAR. **Methods:** Twelve healthy male university students performed 1 repetition maximum (1 RM) tests and an experimental session for 4 consecutive days: sedentary control sessions (Day 1 and 2), high-intensity resistance exercise (bench press) session at 75% of 1 RM (HIRE; Day 3), and recovery session (Day 4). Saliva samples for Days 1-3 were collected: 1) before and 2) immediately after the session, 3) 10 min, 4) 20 min, and 5) 30 min into the recovery period from each session, 6) 9 pm, and 7) 11 pm at night after the session, and 8) immediately after awakening and 9) 15 min and 10) 30 min post-awakening the day after the session (Sampling on Day 4 only upon awakening). **Results:** The results showed that the HIRE session did not change the acute cortisol levels; however, the CAR after the next day showed different responses in those who increased ($n = 6$; Responders) and those who did not increase ($n = 6$; non-Responders). In addition, a significant negative correlation was found between the peak percentage increases in the area under the curve for the whole of CAR (AUC_g) and peak Δ physical fatigue ($r = -.633, p = .027$) and Δ muscle soreness ($r = -.791, p = .002$). **Conclusions:** These results suggest that both increased and no increased CAR may result from physical adaptation and maladaptation to the HIRE session.

Key Words: exercise stress, endocrine, physical adaptation-maladaptation, biomarker

Introduction

Exercise training has positive effects that lead to improved physical functions, such as enhanced energy metabolism and circulation (adaptation), but also negative effects, such as causing overtraining (maladaptation) (Fry et al., 1991). The duality of exercise effects (adaptation-maladaptation) can be explained by the concept of allostasis – the ability to maintain stability through change (McEwen, 1998). When there is a proper exercise and training load and/or recovery period from the load, it functions in allostasis and provides the positive effect of improving physical functions. If these are not effectively performed, the allostatic load, a limiting factor in allostasis, may increase, which results in the negative effect of disruption of physical functions.

Cortisol, which sensitively reflects the activity of the hypothalamic-pituitary-adrenal (HPA) axis, has been studied extensively as one of the evaluation indicators of the allostatic process (Gunnar & Vazquez, 2001; Ockenfels et al., 1995). Cortisol has a diurnal rhythm that is high in the morning and low at night (Allolio et al., 1990) and occurs in acute increases in concentration as a response to stressful stimuli (Dickerson & Kemeny, 2004; Hill et al., 2008; VanBruggen et al., 2011). In addition to these reactions, a phenomenon known as the cortisol awakening response, in which cortisol levels increase during the first 30-45 minutes after awakening, was observed (Pruessner et al., 1997). CAR is generally governed by hypothalamic and pituitary hormone regulation and is estimated to be regulated by recognizing daily demands, and may be critical in giving the necessary energy while switching from a resting to an active state (Pruessner et al., 1997; Stalder et al., 2011).

Recently, CAR has been studied in the context of exercise and training, suggesting that exercise and training load can influence its responses. After a seven-day hard training phase, CAR noticeably increased, and the lower the CAR, the higher the rate of decline in performance among elite soccer players (Minetto et al., 2008). In addition, the study, which compared CAR in athletes diagnosed with overtraining syndrome, healthy athletes, and sedentary controls, reported blunted CAR in athletes diagnosed with overtraining syndrome (Anderson et al., 2021). These results may reflect the progressive process of adrenal disturbance-adaptation-maladaptation, consistent with the “prolonged response” model of allostatic loads since over-or under-activity of the CAR can occur depending on the degree of physical condition (Anderson & Wideman, 2017). Therefore,

there are suggestions that increased and blunted CAR may indicate physical adaptation and maladaptation to exercise and training and may be an early predictor of overtraining.

Since most of the previous studies on CAR were field studies, only a few studies have examined the effects of a single exercise load on CAR; given that the allostatic response results from the accumulation of responses to a single exercise load, the effect of a single exercise load on CAR responses should also be investigated. Several studies have suggested that exercise may have a residual effect that enhances the CAR on the following day. One study has shown that the training load from the previous day had a positive association with CAR (Anderson et al., 2018). In an experimental study with a cycle ergometer, high-intensity exercise resulted in higher CAR on the following day, suggesting that CAR may be influenced by the threshold of exercise intensity on the previous day (Ogasawara et al., 2022). However, exercise and training situations may incorporate resistance exercise to improve muscle hypertrophy and maximal muscle strength into their training program, in addition to aerobic exercise to improve cardiorespiratory endurance. Considering that CAR is used as an indicator to assess physical adaptation-maladaptation to exercise and training loads, it should also examine the effects of resistance exercise.

With these considerations in mind, the purpose of this study was to investigate the effect of a single resistance exercise on CAR. It is generally recognized that single overload resistance exercise causes acute exhaustion and a relative decline in performance (Bell et al., 2020). Thus, the residual effect of a single high-intensity resistance exercise would be expected to result in "over-activity" of the allostatic response on the following day. Therefore, we hypothesized that CAR would increase with high-intensity resistance exercise. However, it has also been reported that there are high individual differences in the CAR response to exercise and training load (Minetto et al., 2008). If the hypotheses were rejected, the analysis was conducted by focusing on individual differences.

Material & methods

Study Design and Setting

This study used strength testing and four consecutive days of experimental sessions to investigate the effects of a single high-intensity resistance exercise on CAR (Fig. 1). The first visit (strength testing) was used for screening and measuring their one-repetition maximum (1RM). The next four days were conducted experimental sessions that continued as follows: two days of sedentary control sessions (Day 1 and Day 2), a high-intensity resistance exercise session until exhaustion (HIRE; Day 3), and a recovery session considering the effect of delayed onset muscle soreness which occurs 8 to 72 hours after exercise (Recovery; Day 4) (Nosaka & Newton, 2002). Strength testing and experimental sessions were separated by at least 72 hours. The CAR was measured during the day after each session, and each response was compared.

Bench press exercise was selected in this study as it has been commonly used among trained individuals, and the load can be easily controlled to ensure safety (Keogh et al., 1999). The intensity of 75% of 1RM was selected because it is quite commonly used in many resistance training programs (Kraemer & Ratamess, 2004).

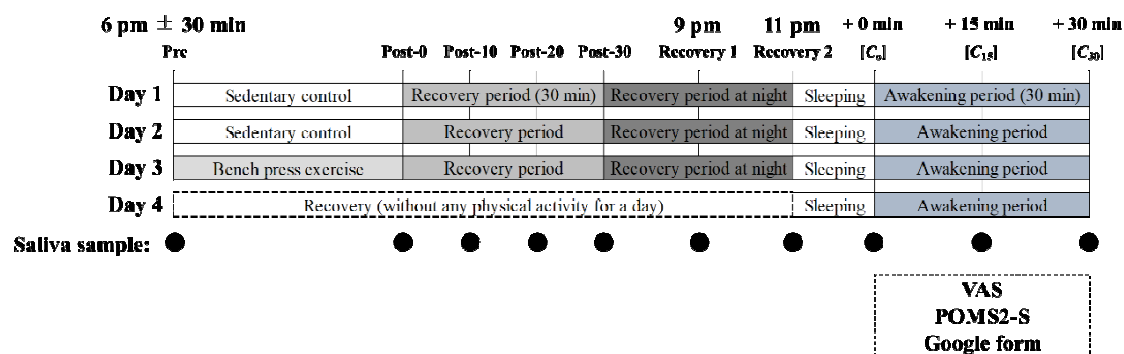


Fig. 1. Study protocol

Participants

Before the experiment, all participants were provided with oral and written informed consent. Fourteen healthy male university students with at least 1 year of resistance exercise experience participated in this study (mean ± SD; 21.9 ± 0.9 years, 171.2 ± 5.8 cm, 68.3 ± 6.0 kg, 23.3 ± 1.4 BMI, 88.08 ± 17.8 kg bench press 1RM). Exclusion criteria included a history of hormonal disorders, mental illness, smoking, a diet chronically low in carbohydrates, use of anabolic steroids, or chronic nonsteroidal anti-inflammatory drug use. Two participants were excluded since one was affected by other exercises, and the other had blunted CAR throughout

the experimental period. Therefore, 12 participants' data were used for further analyses. This research was approved by the Ethics Committee at Osaka University of Health and Sport Sciences (approval number: 20-19).

Procedures

Strength Testing.

At least 72 hours prior to the experimental session, each participant had their 1 RM determined on the bench press exercise. The 1 RM tests were conducted using the National Strength and Conditioning Association protocol for 1 RM testing (Jeremy & Triple, 2015). The protocol required the participants to progressively increase the resistance across several trials until the 1 RM was achieved. For the lift to be considered successful, each participant had to begin by holding the barbell in the traditional shoulder-width grip with their elbows fully extended. The barbell was then lowered until it lightly touched each participant's chest before being returned to the starting position. The participants were not allowed to bounce the bar off their chests at any point during the exercise. Participants were expected to keep contact between their feet and the floor during the entirety of the lift and their hips, shoulders, and head with the bench (Hoffman, 2006). Additionally, they were instructed to raise their maximum speed for each of the trials.

Experimental Session.

On Day 1 and Day 2, a sedentary control session (20 minutes) was conducted to measure their baseline values. On Day 3, the participants performed bench press exercises with (a) 20 kg for 10 repetitions and (b) 40% 1 RM for 10 repetitions as a warm-up exercise. After the warm-up, participants completed the HIRE session until exhaustion, which consisted of 5-8 sets of up to 10 repetitions at 75% 1RM with a 90-second rest between sets. Participants were instructed to complete as many repetitions as possible until either reaching 10 repetitions or muscular failure in each set. Failure was defined as the inability to complete a full repetition without assistance (Gonzalez et al., 2018). Rating of perceived exertion (RPE) was recorded after each set of the barbell bench press exercise, as well as after the entire HIRE session, and sessions were continued until the RPE was 19 or greater. On Day 4, a Recovery session without any physical activity for a day was conducted for an additional day after the HIRE session.

As shown in Fig. 1, saliva samples for the control and the HIRE sessions were taken based on previous research (Ogasawara et al., 2022); before (Pre) and immediately after (Post-0) the experimental session, during the recovery period of the session (every 10 min for 30 min; Post-10, Post-20, and Post-30), the night after returning home from the session (9 pm, 11 pm; Recovery 1 and Recovery 2), and upon awakening (immediately after awakening and 15 min and 30 min post-awakening) the day after the session. In addition, perceived parameters were measured upon awakening to understand the factors for changes in CAR. The Recovery session of Day 4 was conducted only to measure upon awakening the next day. Participants started all sessions at the same time of the day (6 pm \pm 30 min) to control for biological variation. They were asked to eat similar meals during the experiment and to refrain from eating at least 4 hours before the experiment and after 8 pm after the experiment. In addition, the participants were required to refrain from exercise, caffeine, and alcohol during the four-day experiment.

Instruments

Rating of Perceived Exertion.

The rating of perceived exertion (RPE) was determined using Borg's 6-20 points rating of perceived exertion scale, a validated Japanese questionnaire (Onodera & Miyashita, 1976).

Cortisol measurements.

Saliva samples were collected (Fig. 1) by the passive drool techniques. Prior to the experiment, the participants were informed of the guidelines for saliva collection. Participants collected saliva in their mouths for 2 minutes before pouring it into a tube using a straw. To avoid saliva dilution, no mouthwash was utilized. Participants were asked not to drink, wash their teeth, or bathe for 1 hour before saliva collection for Recovery 1 and 2. During the wakeup phase, they were also not allowed to eat, drink, brush their teeth, engage in vigorous physical activity, or return to sleep, but they were allowed to engage in other normal morning activities. The participants were asked to report their sleeping time, waking time, saliva collection time, and sleeping status on a Google form. The collected saliva was stored at 4°C in a commercial refrigerator and submitted to the laboratory within 48 hours. To precipitate particulate particles, the saliva samples were centrifuged at room temperature for 5 minutes at 3000g and kept at -80°C until analysis. Following that, cortisol concentrations were determined using an enzyme-linked immunosorbent assay (Cortisol ELISA Kit (RE52611), IBL, Japan).

CAR data were also summarized using the Area Under the Curve computation concerning the ground of cortisol levels (AUC_g) (Pruessner et al., 1997). AUC_g were significantly associated between the 2-day sedentary control sessions (Day 1 and Day 2) ($r = .801$, $p = .002$); thus, the mean of Day 1 and Day 2 was used as the Control value (Control). However, Day 1 data for three participants were excluded - one with higher perceived

stress and one with higher physical fatigue compared to the other days, and one with a 10-minute delay in saliva collection upon awakening - and only Day 2 data demonstrated better compliances were used.

Perceptual parameters measured upon awakening.

To explore the factors that changed the CAR resulting from the HIRE session, physical conditions (perceived physical fatigue and muscle soreness of the upper limb) were assessed using a visual analog scale (VAS). In addition, to excluding confounding factors, factors that could influence CAR were measured upon awakening. The degree of stress and sleep quality were evaluated by VAS. Participants were asked to rate their perceived parameters upon awakening by marking the corresponding 10-cm lines.

The psychological condition was assessed with a total mood disturbance (TMD) score calculated from the short form of the Profile of Moods States Second Edition (POMS2-S), which has been translated into Japanese (Yokoyama & Watanabe, 2015). Participants graded a set of 35 items related to the mood on a Likert scale from 0 (not at all) to 4 (extremely) to such question "How do you feel at this moment?" to assess 7 dimensions: Anger-Hostility (AH), Confusion-Bewilderment (CB), Depression-Dejection (DD), Fatigue-Inertia (FI), Tension-Anxiety (TA), Vigor-Activity (VA), and Friendliness (F).

Data Analysis

All the data of control value (Control) were used for the mean of the 2-day sedentary control session (Day 1 and Day 2). The Day 1 data of the three participants excluded for the cortisol data were also excluded in the other data. All data were expressed as mean \pm standard error (SE).

In this study, a statistical examination was conducted in two phases. First, an analysis to examine the effects of the HIRE sessions on the CAR in the overall participants was performed. Two-way repeated-measures ANOVA tests were conducted to analyze the changes in cortisol concentrations on the day of the experimental session (session [2] \times sampling point [7]) and CAR on the day after the session (session [3] \times sampling point [3]). One-way repeated measure ANOVAs were used to analyze the difference between the sessions in VAS (physical fatigue, muscle soreness, stress, and sleep quality), the TMD score during awakening, sleeping hours, and awakening time.

Next, we conducted an analysis to consider individual variability. One-way repeated measure ANOVAs were used to analyze the difference between experimental sessions in AUC_g . After that, the percentage of change in AUC_g from Control to HIRE and Recovery sessions were calculated. Additionally, the median of the peak percentage increase in AUC_g was calculated, and the participants were separated into two groups (Responders: $n = 6$; non-Responders: $n = 6$ group) based on the median (19.4 %). Then, two-way repeated-measures ANOVA tests were performed to analyze AUC_g (group [2] \times session [3]) again. In order to examine the factors that influenced the change in CAR in each individual, we performed correlation analyses (Pearson correlation) to assess the relationship between the peak percentage increase in AUC_g (peak AUC_g , % Control) and the peak absolute increment in physical fatigue (peak Δ physical fatigue) and muscle soreness (peak Δ muscle soreness).

In ANOVA tests, Mauchly's test was used to assess the sphericity assumption. If the assumption was violated, Greenhouse-Geisser epsilon values were used to adjust the degrees of freedom. Bonferroni corrections were used for post hoc tests. The estimate of effect size was quantified by partial eta squared (η_p^2), where $\eta_p^2 = 0.01, 0.06,$ and 0.14 were estimated for a small, moderate, and large effect, respectively (Larson-Hall, 2009). The level of significance was set to $\alpha \leq .05$. In addition, a more liberal α value of $\leq .10$ was used to determine marginal significance to avoid Type II error (Cohen, 1988). All statistical analyses were performed using SPSS version 27.0 (IBM, Japan).

Results

During the HIRE session, the number of repetitions decreased with each set, and finally, the participants completed 41.4 ± 11.1 repetitions of lifting. In addition, the RPE value at the last repetition was 19.8 ± 0.1 .

Cortisol Data Throughout the Experimental Session

The changes in salivary cortisol concentration on the day of the experimental session and CAR on the day after the experimental session are shown in Fig. 2.

Salivary cortisol concentration on the day of the experimental session showed that the main effect was significant at sampling point ($F(6, 66) = 9.099, p = .004, \epsilon = .255, \eta_p^2 = .453$). The main effect of session ($F(1, 11) = 0.280, p = .607, \eta_p^2 = .025$) and the interaction between session and sampling point ($F(6, 66) = 0.976, p = .384, \epsilon = .290, \eta_p^2 = .081$) were not significant. The results of multiple comparisons at the sampling points were lower for Recovery 2 compared to Pre ($p = .07$).

CAR on the day after the experimental session showed that the main effect was significant at sampling point ($F(2, 22) = 18.062, p = .001, \eta_p^2 = .621$). The main effect of session ($F(2, 22) = 0.518, p = .365, \epsilon = .635, \eta_p^2 = .032$) and the interaction between session and sampling point ($F(4, 44) = 1.437, p = .259, \epsilon = .505, \eta_p^2 = .116$) were not significant. The results of multiple comparisons at the sampling points showed a significant increase from C_0 to C_{30} ($C_0 < C_{15} < C_{30}; p < .05$, respectively).

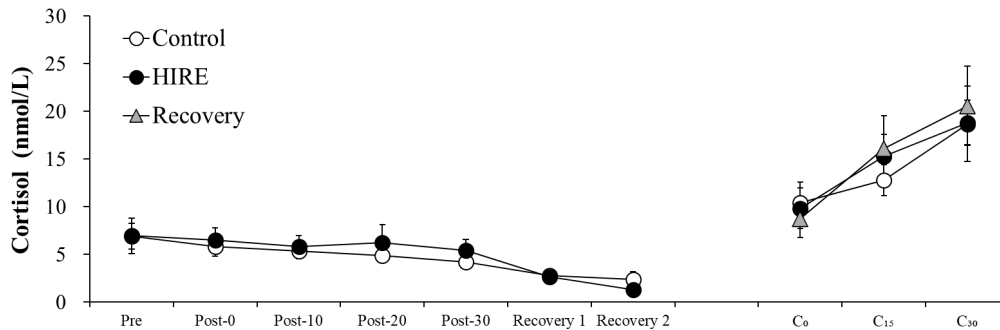


Fig. 2. Cortisol data throughout the experimental session ($M \pm SE$)

Factors that Influenced the CAR

Factors that influenced the CAR measured upon awakening are shown in Table 1 and Table 2. The main effect was significant for muscle soreness ($F(2, 22) = 13.779, p = .001, \eta_p^2 = .556$) and a significant tendency for physical fatigue ($F(2, 22) = 3.403, p = .052, \eta_p^2 = .236$) at session. Post-hoc tests of muscle soreness confirmed that it was significantly higher in the HIRE session and Recovery session than in Control ($p = .002, p = .001$, respectively). Post-hoc tests of physical fatigue confirmed that it was higher in the Recovery session than in Control ($p = .091$). There were no other significant main effects.

Table 1. Factors influencing CAR as measured at awakening ($M \pm SE$)

| Measure | Control | HIRE | Recovery |
|------------------|------------|-------------|-------------------------|
| Physical fatigue | 11.6 ± 2.8 | 23.4 ± 5.3 | 26.4 ± 5.5 [†] |
| Muscle soreness | 3.5 ± 1.7 | 37.0 ± 7.4* | 37.4 ± 5.0* |
| Stress | 14.7 ± 3.1 | 13.5 ± 2.9 | 19.7 ± 5.1 |
| Sleep quality | 64.3 ± 5.7 | 68.2 ± 6.6 | 56.7 ± 7.9 |
| TMD score | 97.9 ± 3.4 | 96.8 ± 3.4 | 97.5 ± 3.5 |

Physical fatigue was higher on Recovery than on Control ([†] $p < .10$)

Muscle soreness was significantly higher on HIRE and Recovery than on Control (* $p < .05$).

Table 2. Sleeping hours, awakening time, and first sampling point during the experimental session ($M \pm SE$)

| Measure | Control | HIRE | Recovery |
|---------------------|-------------|-------------|-------------|
| Sleeping Hours (h) | 6.9 ± 0.4 | 6.9 ± 0.3 | 6.9 ± 0.4 |
| Awakening Time | 7:46 ± 0:24 | 8:15 ± 0:20 | 7:57 ± 0:29 |
| First Sampling Time | 7:48 ± 0:25 | 8:16 ± 0:20 | 7:59 ± 0:29 |

The Result of Considering Individual Adaptation

AUC_g data of each participant can be seen in Fig. 3 A). The main effect of session was not significant ($F(2, 22) = .668, p = .461, \epsilon = .626, \eta_p^2 = .057$), but the individual data appeared to show different adaptations in each participant. After grouping the results (Fig. 3 B)), the interaction between group and session was significant ($F(2, 20) = 4.668, p = .047, \epsilon = .597, \eta_p^2 = .318$). The main effect of group showed a significant tendency ($F(1, 10) = 3.771, p = .081, \eta_p^2 = .274$).

The main effect of session was not significant ($F(2, 22) = 0.890, p = .426, \epsilon = .597, \eta_p^2 = .082$). Post-hoc tests of group confirmed that the Responders group showed higher values at the HIRE session ($p = .089$) and significantly higher values at the Recovery session ($p = .044$) compared to the non-Responders group. Post-hoc tests of session confirmed that the HIRE and the Recovery sessions were higher than Control in the Responders group ($p < .05, p < .10$, respectively).

There were no significant differences between sessions in the non-Responders group.

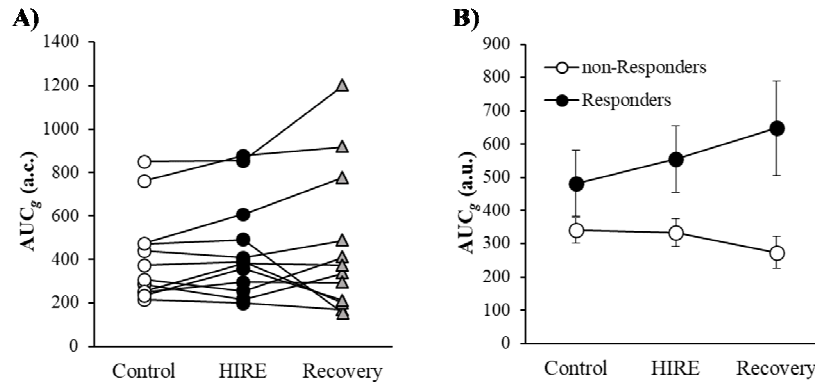


Fig. 3. A) Individual AUC_g data and B) mean ($\pm SE$) levels of AUC_g in Responders and non-Responders.

The correlation analysis showed a significant negative relationship between peak AUC_g , % Control and peak Δ physical fatigue ($r = -.633, p = .027$) and peak Δ muscle soreness ($r = -.791, p = .002$) (Fig. 4).

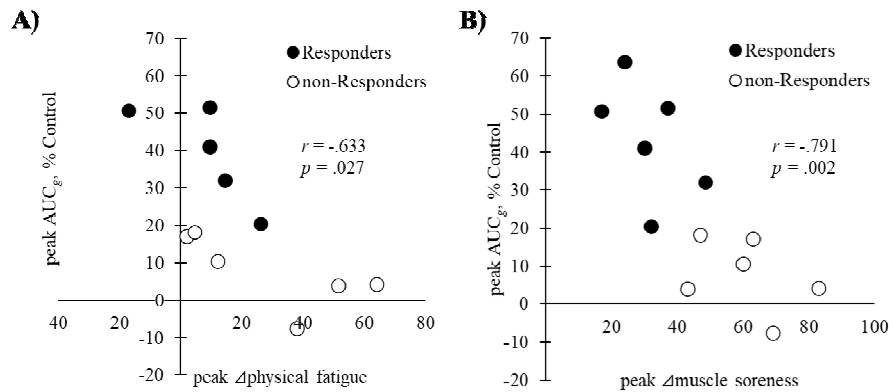


Fig. 4. Relationship between peak AUC_g , % Control and A) peak Δ physical fatigue and B) peak Δ muscle soreness. The horizontal axis shows the peak increase in physical fatigue and muscle soreness, respectively, and the vertical axis shows the peak percentage increase in AUC_g .

Discussion

This study aimed to investigate the effect of a single high-intensity resistance exercise on CAR in healthy participants. Our results showed that the HIRE session did not change the cortisol concentration on the day of the experiment; however, the CAR showed different responses in those who increased and did not increase. In addition, a significant negative correlation was found between peak AUC_g , % Control and peak Δ physical fatigue, and peak Δ muscle soreness. These results suggest that an increase and no increase of CAR may result from physical adaptation and maladaptation to the HIRE sessions. This was the first study to demonstrate the response of CAR to a single experimental resistance exercise.

All participants performed the HIRE sessions to exhaustion. This was clear from the results of the RPE. Nevertheless, salivary cortisol levels did not change in the HIRE session. This result was different from our previous study with a cycle ergometer (Ogasawara et al., 2022). The metabolic stimulation of cortisol secretion was reported to be less prominent in the resistance exercise paradigm than in endurance exercise (Anderson et al., 2019). Therefore, the acute effects of resistance exercise on the HPA axis are not typically as pronounced. Training intensities aimed at improving muscle strength/power (88% of 1 RM) did not produce acute changes in cortisol levels (Crewther et al., 2008; Smilios et al., 2003).

On the other hand, metabolic demanding higher total work protocols (i.e., high volume, moderate to high intensity with short rest periods) elicited a greater acute cortisol response as part of a larger remodeling process in muscle tissue, even in the context of resistance exercise (Kraemer & Ratamess, 2005). Training intensities aimed at improving muscle hypertrophy (75% of 1RM) and muscular endurance (60% of 1RM) resulted in acute increases in cortisol concentrations (Crewther et al., 2008; Smilios et al., 2003). The HIRE sessions in this study were performed at the intensity of 75% of 1RM but did not increase the cortisol levels. In the above previous study, the participants performed exercises that mobilized larger amounts of muscle mass (squat lifts, full-body exercises combining multiple disciplines) compared to the bench press exercises performed in this study. Therefore, the bench press exercise, which mobilizes a small amount of muscle, might have been a

factor that did not cause an acute cortisol response. The decreased level of cortisol from pre to Recovery 2 was perhaps not because of resistance exercise but rather due to the diurnal variation of cortisol, which was high in the morning and low at night (Allolio et al., 1990).

The results of CAR on the day after the experimental session showed no change for overall participants; however, there were two types of participants at the individual level: the ones whose CAR increased (Responders) and the others whose CAR did not increase (non-Responders). There was no significant difference in the number of repetitions completed between the Responders and non-Responders groups (Responders group: 38.2 ± 12.7 ; non-Responders group: 44.7 ± 8.0), and there were no differences in perceived parameters (except for physical fatigue and muscle soreness) that could influence the CAR (data not reported). Thus, differences in CAR responses to the HIRE session among participants may be a major reason this study's results did not support the hypothesis. It is generally considered that resting cortisol concentrations reflect the adaptation to chronic training stress (Kraemer & Ratamess, 2005), but chronic resistance exercise did not produce consistent cortisol secretion. There have been reports of unchanged (Häkkinen et al., 1987, 2000), decreased (Alen et al., 1988; Kraemer et al., 1998), and increased (Häkkinen & Pakarinen, 1991) resting cortisol concentrations during normal strength and power training in men and women during short-term overreaching, respectively. In the context of the CAR study, there were also participants whose CAR increased and did not increase (or even decrease) during the 7-day strength training period (Minetto et al., 2008). In the study, the rate of decrease in performance was positively correlated with the respective CAR data, suggesting that increased CAR may be the result of training load-induced hyper-responsiveness of the HPA axis.

In contrast, unchanged and decreased CAR might reflect dysregulated adaptation to training load due to a subclinical form of hypocortisolism. In this study, the HIRE session caused physical fatigue and delayed onset muscle soreness, and a negative correlation was found between peak AUC_g , % Control and peak Δ physical fatigue, and peak Δ muscle soreness. Single overload resistance exercise caused acute exhaustion and a relative decline in performance (Bell et al., 2020). Considering the association with physical fatigue and delayed onset muscle soreness, the increase and unchanged/decrease in CAR observed in this study may indicate temporary hyper-responsiveness and dysregulation of the HPA axis, as shown in previous studies (Minetto et al., 2008). However, changes in performance were not measured in this study. Thus, the physical adaptation or maladaptation to the training load could not be completely concluded from the response of CAR in this study.

The limitations of the present study should be noted. First, the small sample size makes it difficult to generalize the findings. In particular, a larger sample size would be needed to generalize the results for Responders and non-Responders groups. Second, the lack of measurement of the participants' performance prevents us from completely concluding the relationship between changes in CAR and physical adaptation. The increased and unchanged CAR observed in the present study may be interpreted as at a point in time on the "prolonged response" model of allostatic load proposed by McEwen (1998), as in the previous study (Minetto et al., 2008). However, making such a conclusion from only the relationship between changes in CAR and subjective evaluations would be difficult. Finally, it is interesting to note that CAR responded the day after the HIRE session even though an acute cortisol response did not occur. Future studies would be expected to examine the mechanism using other metabolic response markers and the effects of other resistance exercises that mobilized larger amounts of muscle mass or different intensity resistance exercises based on the total workload of the training.

Conclusions

In the present study, CAR increased and/or unchanged (or even decrease) following high-intensity resistance exercise, and a negative correlation was found between the CAR response and physical fatigue and muscle soreness. Although an issue of evaluating CAR responses along with performance remains, the results suggest that CAR has the potential to assess short-term physical adaptation to high-intensity resistance exercise. This finding may help individuals involved in training plan future training loads and recovery.

Conflicts of interest

There is no conflict of interest to declare.

Acknowledgments

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References

- Alen, M., Pakarinen, A., Häkkinen, K., & Komi, P. V. (1988). Responses of serum androgenic-anabolic and catabolic hormones to prolonged strength training. *Int J Sports Med*, 9(3), 229-233. <https://doi.org/10.1055/s-2007-1025011>
- Allolio, B., Hoffmann, J., Linton, E. A., Winkelmann, W., Kusche, M., & Schulte, H. M. (1990). Diurnal salivary cortisol patterns during pregnancy and after delivery: relationship to plasma corticotrophin-releasing-hormone. *Clin Endocrinol (Oxf)*, 33(2), 279-289. <https://doi.org/10.1111/j.1365-2265.1990.tb00492.x>

- Anderson, T., & Wideman, L. (2017). Exercise and the cortisol awakening response: a systematic review. *Sports Med Open*, 3(1), 1-15. <https://doi.org/10.1186/s40798-017-0102-3>
- Anderson, T., Berry, N. T., & Wideman, L. (2019). Exercise and the hypothalamic-pituitary-adrenal axis: a special focus on acute cortisol and growth hormone responses. *Current Opinion in Endocrine and Metabolic Research*, 9, 74-77. <https://doi.org/10.1016/j.coemr.2019.08.002>
- Anderson, T., Lane, A. R., & Hackney, A. C. (2018). The cortisol awakening response: association with training load in endurance runners. *Int J Sports Physiol Perform*, 13(9), 1158-1163. <https://doi.org/10.1123/ijsp.2017-0740>
- Anderson, T., Wideman, L., Cadegiani, F. A., & Kater, C. E. (2021). Effects of overtraining status on the cortisol awakening response — endocrine and metabolic responses on overtraining syndrome (EROS-CAR). *Int J Sports Physiol Perform*, 16(7), 965-973. <https://doi.org/10.1123/ijsp.2020-0205>
- Bell, L., Ruddock, A., Maden-Wilkinson, T., & Rogerson, D. (2020). Overreaching and overtraining in strength sports and resistance training: a scoping review. *J Sports Sci*, 38(16), 1897-1912. <https://doi.org/10.1080/02640414.2020.1763077>
- Cohen, J. (1988). *Statistical power analysis for the behavioral sciences* (2nd ed.). Routledge. <https://doi.org/10.4324/9780203771587>
- Crewther, B., Cronin, J., Keogh, J., & Cook, C. (2008). The salivary testosterone and cortisol response to three loading schemes. *J Strength Cond Res*, 22(1), 250-255. <https://doi.org/10.1519/JSC.0b013e31815f5f91>
- Dickerson, S. S., & Kemeny, M. E. (2004). Acute stressors and cortisol responses: a theoretical integration and synthesis of laboratory research. *Psychol Bull*, 130(3), 355-391. <https://doi.org/10.1037/0033-2909.130.3.355>
- Fry, R. W., Morton, A. R., & Keast, D. (1991). Overtraining in athletes. An Update. *Sports Med*, 12(1), 32-65. <https://doi.org/10.2165/00007256-199112010-00004>
- Gonzalez, A. M., Spitz, R. W., Ghigiarelli, J. J., Sell, K. M., & Mangine, G. T. (2018). Acute effect of citrulline malate supplementation on upper-body resistance exercise performance in recreationally resistance-trained men. *J Strength Cond Res*, 32(11), 3088-3094. <https://doi.org/10.1519/JSC.0000000000002373>
- Gunnar, M. R., & Vazquez, D. M. (2001). Low cortisol and a flattening of expected daytime rhythm: potential indices of risk in human development. *Dev Psychopathol*, 13(3), 515-538. <https://doi.org/10.1017/S0954579401003066>
- Häkkinen, K., & Pakarinen, A. (1991). Serum hormones in male strength athletes during intensive short term strength training. *Eur J Appl Physiol*, 63(3), 194-199. <https://doi.org/10.1007/BF00233847>
- Häkkinen, K., Pakarinen, A., Alen, M., Kauhanen, H., & Komi, P. V. (1987). Relationships between training volume, physical performance capacity, and serum hormone concentrations during prolonged training in elite weight lifters. *Int J Sports Med*, 8, S61-S65. <https://doi.org/10.1055/s-2008-1025705>
- Häkkinen, K., Pakarinen, A., Kraemer, W. J., Newton, R. U., & Alen, M. (2000). Basal concentrations and acute responses of serum hormones and strength development during heavy resistance training in middle-aged and elderly men and women. *J Gerontol A Biol Sci Med Sci*, 55(2), B95-105. <https://doi.org/10.1093/gerona/55.2.B95>
- Hill, E. E., Zack, E., Battaglini, C., Viru, M., Viru, A., & Hackney, A. C. (2008). Exercise and circulating cortisol levels: the intensity threshold effect. *J Endocrinol Invest*, 31(7), 587-591. <https://doi.org/10.1007/BF03345606>
- Hoffman, J. R. (2006). Muscular Strength. In *Norms for Fitness, Performance, and Health*. (pp. 32-36). Human Kinetics.
- Jeremy, M. S., Triplet, N. T. (2015). Program design for resistance training. In Haff, G. G., & Triplett, N. T. (Eds.), *Essentials of strength training and conditioning* (4th ed.). (pp. 439-470). Human Kinetics.
- Keogh, J. W., Wilson, G. J., & Weatherby, R. E. (1999). A cross-sectional comparison of different resistance training techniques in the bench press. *J Strength Cond Res*, 13(3), 247-258. Retrieved from <https://journals.lww.com/nsca-jscr/pages/default.aspx>
- Kraemer, W. J., & Ratamess, N. A. (2004). Fundamentals of resistance training: progression and exercise prescription. *Med Sci Sports Exerc*, 36, 674-688. <https://doi.org/10.1249/01.mss.0000121945.36635.61>
- Kraemer, W. J., & Ratamess, N. A. (2005). Hormonal responses and adaptations to resistance exercise and training. *Sports Med*, 35(4), 339-361. <https://doi.org/10.2165/00007256-200535040-00004>
- Kraemer, W. J., Staron, R. S., Hagerman, F. C., Hikida, R. S., Fry, A. C., Gordon, S. E., Nindl, B. C., Gothshalk, L. A., Volek, J. S., Marx, J. O., Newton, R. U., & Häkkinen, K. (1998). The effects of short-term resistance training on endocrine function in men and women. *Eur J Appl Physiol*, 78(1), 69-76. <https://doi.org/10.1007/s004210050389>
- Larson-Hall, J. (2009). *A guide to doing statistics in second language research using SPSS*. Routledge. <https://doi.org/10.4324/9781315775661>
- McEwen, B. S. (1998). Protective and damaging effects of stress mediators. *N Engl J Med*, 338(3), 171-179. <https://doi.org/10.1056/NEJM199801153380307>

- Minetto, M. A., Lanfranco, F., Tibaudi, A., Baldi, M., Termine, A., & Ghigo, E. (2008). Changes in awakening cortisol response and midnight salivary cortisol are sensitive markers of strenuous training-induced fatigue. *J Endocrinol Invest*, *31*(1), 16-24. <https://doi.org/10.1007/BF03345561>
- Nosaka, K., & Newton, M. (2002). Is recovery from muscle damage retarded by a subsequent bout of eccentric exercise inducing larger decreases in force? *J Sci Med Sport*, *5*(3), 204-218. [https://doi.org/10.1016/S1440-2440\(02\)80005-6](https://doi.org/10.1016/S1440-2440(02)80005-6)
- Ockenfels, M. C., Porter, L., Smyth, J., Kirschbaum, C., Hellhammer, D. H., & Stone, A. A. (1995). Effect of chronic stress associated with unemployment on salivary cortisol: overall cortisol levels, diurnal rhythm, and acute stress reactivity. *Psychosom Med*, *57*(5), 460-467. <https://doi.org/10.1097/00006842-199509000-00008>
- Ogasawara, Y., Kadooka, S., Tsuchiya, H., & Sugo, T. (2022). High cortisol awakening response measured on the day following high-intensity exercise. *J Phys Fitness Sports Med*, *11*(2), 59-66. <https://doi.org/10.7600/jpfsfm.11.59>
- Onodera, K., & Miyashita, M. (1976). A study on Japanese scale for rating of perceived exertion in endurance exercise [in Japanese]. *Taiikugaku Kenkyu (Japan Journal of Physical Education, Health and Sport Sciences)*, *21*(4), 191-203. <https://doi.org/10.5432/jjpehss.KJ00003405473>
- Pruessner, J. C., Wolf, O. T., Hellhammer, D. H., Buske-Kirschbaum, A., von Auer, K., Jobst, S., Kaspers, F., & Kirschbaum, C. (1997). Free cortisol levels after awakening: a reliable biological marker for the assessment of adrenocortical activity. *Life Sci*, *61*(26), 2539-2549. [https://doi.org/10.1016/S0024-3205\(97\)01008-4](https://doi.org/10.1016/S0024-3205(97)01008-4)
- Smiliotis, I., Piliandis, T., Karamouzis, M., & Tokmakidis, S. P. (2003). Hormonal responses after various resistance exercise protocols. *Med Sci Sports Exerc*, *35*(4), 644-654. <https://doi.org/10.1249/01.MSS.0000058366.04460.5F>
- Stalder, T., Evans, P., Hucklebridge, F., & Clow, A. (2011). Associations between the cortisol awakening response and heart rate variability. *Psychoneuroendocrinology*, *36*(4), 454-462. <https://doi.org/10.1016/j.psyneuen.2010.07.020>
- VanBruggen, M. D., Hackney, A. C., McMurray, R. G., & Ondrak, K. S. (2011). The relationship between serum and salivary cortisol levels in response to different intensities of exercise. *Int J Sports Physiol Perform*, *6*(3), 396-407. <https://doi.org/10.1123/ijsp.6.3.396>
- Yokoyama, K., & Watanabe, K. (2015). *Profile of mood states* (2nd ed.). Kaneko Shobo.