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**Original Article**

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# Effects of an incremental maximal endurance exercise stress-induced cortisol on cognitive performance

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## ABSTRACT

**Objectives:** It can be hypothesized that cognitive performance decreases after fatigue protocol when it coincides with the maximum peak of cortisol. The first aim of this study was to elucidate the effects of a single bout of high intensity exercise on behavioural (i.e., attention and memory) and physiological (i.e., salivary cortisol) responses. The second objective was to evaluate the effect of the performance of the cognitive tasks on cortisol levels. **Methods:** Thirty-four physically active men (at least 5 days/week of physical activity practice) 38.11 (1.57) years old completed a maximal incremental protocol on a treadmill by running until they reached a state of stress. Salivary cortisol and cognitive functions were evaluated in counterbalanced order prior and following exercise-induced stress. **Results:** Results showed lower cortisol levels before exercise and higher cortisol values before the cognitive task. Indeed, exercise-induced stress had only a detrimental effect on attention without any impact on declarative memory and finding improvements on working memory performance. **Conclusion:** The effects of stress on cognitive performance depending on the main brain areas responsible of cognitive functions (i.e., prefrontal cortex and hippocampus) and time

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elapsed between the cessation of exercise and the evaluation of these. **Keywords:** Exercise; Stress; Fatigue; Cognitive functions; Cortisol.

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## INTRODUCTION

The exercise is well known for activating the body's stress response, the hypothalamic-pituitary-adrenal (HPA) axis, producing effects at a physical and psychological level (Heijnen, Hommel, Kibele, & Colzato, 2016). One of these physiological responses is the secretion of the final product of the HPA axis: cortisol.

Cortisol is the main glucocorticoid hormone, uniting around 95% of the glucocorticoid activity in humans (Stoelting & Hillier, 2012) and acts as a trigger of the homeostatic defence mechanisms in the organism (Nicolson, 2008). Blood and salivary cortisol concentrations are commonly used as indicators of the psycho-neuro-endocrinal response to stress (Duclos et al., 1998; Labsy et al., 2013) and both parameters are closely correlated (Vining & McGinley, 1987).

The effects of cortisol on different cognitive functions (e.g., inhibitive control, attention, and memory) have been previously studied (Dickerson & Kemeny, 2004; Henckens, Wingen, A, Joëls, & Fernández, 2012; van Ast et al., 2013). However, empirical findings have shown controversial findings, i.e., while some studies have reported a positive effect of increased cortisol levels on cognitive performance (Bourne Jr & Yaroush, 2003; Cioncoloni et al., 2014), conversely, other studies have reported that increased stress causes different levels of degradation in cognitive functioning, proving that the person makes mistakes or makes inappropriate responses (Gaydos, Curry, & Bushby, 2013; van Enkhuizen et al., 2014). Other authors have argued that the effects of stress in cognition often follow an inverted U-shaped curve (Joëls, Pu, Wiegert, Oitzl, & Krugers, 2006), while moderate levels of stress often result in positive effects on cognitive performance (Guenzel, Wolf, & Schwabe, 2014; Wolf, 2009), high increases produce a significant reduction in cognitive performance (Butler, Klaus, Edwards, & Pennington, 2017; Morgan, Doran, Steffian, Hazlett, & Southwick, 2006; Morgan et al., 2011; Veld, Riksen-Walraven, & Weerth, 2014). However, due to the contradictory findings this issue remains unclear.

Regarding the role of acute exercise in cognitive functioning (Heijnen et al., 2016), some studies have found that aerobic endurance exercise increases the warning state and neural activation of the participants (Y. K. Chang, Labban, Gapin, & Etnier, 2012; Y.-K. Chang, Tsai, Huang, Wang, & Chu, 2014; Dietrich & Audiffren, 2011; Lambourne & Tomporowski, 2010; Magnié et al., 2000) suggesting that acute bouts of exercise may selectively boost executive function performance involving inhibitory control and attention (Drollette, Shishido, Pontifex, & Hillman, 2012; Tsai et al., 2014). In addition, previous research has shown that acute aerobic exercise reduces reaction times (RT) (Audiffren, Tomporowski, & Zagrodnik, 2008; Bermejo, García-Massó, Paillard, & Noé, 2018; Hsieh, Chang, Hung, & Fang, 2016) although this facilitation does not seem to extend into the post-exercise period more a few minutes (Dietrich & Audiffren, 2011) and may even be reversed into an impairment when exercise is prolonged and/or reaches exhaustion (Cian, Barraud, Melin, & Raphael, 2001; Sudo et al., 2017).

This disparity of results does not necessarily question the assumption that there is a positive relationship between acute exercise and cognition, but rather, shows that this relationship is complex and sensitive to multiple factors (Tomporowski, Davis, Miller, & Naglieri, 2008). For example, the time gap between the cessation of exercise and the evaluation of cognitive functioning is one crucial variable due to the transience of the psychophysiological effects of the acute exercise (Crabbe & Dishman, 2004). Therefore, considering that intense exercise may be considered a stressor (Tsai et al., 2014), and consequently modulate the performance of cognitive functions (Lambourne & Tomporowski, 2010) and cortisol levels (Engel et al., 2014), the latter could be related to the effects of exercise on cognition (Henckens et al., 2012).

Indeed, according to some authors the beneficial effects of a single bout of acute exercise on cognitive performance could be related to acute decreases in cortisol levels (Heaney, Carroll, & Phillips, 2013), while detrimental effects on such cognitive functions could, to some extent, be due to a pronounced cortisol stress response (Almela, van der Meij, Hidalgo, Villada, & Salvador, 2012; Quesada, Wiemers, Schoofs, & Wolf, 2012).

Taking all this into account, the concurrent effect of acute exercise on cognitive functioning and the endocrinological system have yielded inconsistent results. Therefore, the first aim of this study was to elucidate the effects of a single bout of high intensity exercise on behavioural (i.e., attention and memory) and physiological (i.e., salivary cortisol) responses. The second objective was to evaluate the effect of the performance of the cognitive tasks on cortisol levels. It can be hypothesized that cognitive performance decreases after fatigue protocol when it coincides with the maximum peak of cortisol.

## METHODS

### *Participants*

Thirty-four males participated voluntarily in this study. The age of the participants was between 24 and 48 years old [mean (SE); 38.11 (1.57) years old, 171.74 (2.36) cm of height and 78.08 (1.56) kg of weight]. All subjects were physically active (at least 5 days/week of physical activity practice). Exclusion criteria were: neurological disorders, sensory disorders or medication that might influence their results.

Subjects provided informed consent before participating in the study. The protocols used in this research work received ethical clearance by the University of Valencia's Ethical Committee. These protocols also met the requirements set out in the Declaration of Helsinki, 1975, which was subsequently reviewed in 2008.

### *Overall procedure*

Before the experimental session, subjects performed a familiarization session with the tasks included in the study. Moreover, subjects were instructed by researchers not to take stimulants (e.g., coffee) nor perform exercise 24 hours before the study.

One week after the familiarization session, the subjects performed the experimental session during which data was acquired. Only one experimental session was completed. In order to control for circadian cortisol variations, all testing took place in the afternoon between 13:30 and 18:00 h (Nicolson, 2008). Once subjects arrived at the laboratory, they were fitted with a Polar RS800CX HR monitor (Polar Electro Ltd., Kempele, Finland). Subsequently, the subjects passed a relaxation phase where they lay on a stretcher and were following the pace them a metronome marking of 40 beats/min with the breath. Cortisol baseline measurements ( $C_1$ ) were obtained after a relaxation phase of approximately 15 min. Then, cognitive performance of the subjects was evaluated with two tasks: (i) Digit Span (DS) for working memory and executive function (Wechsler et al., 1997), and (ii) Psychomotor Vigilance Task (PVT) for alertness (Basner & Dinges, 2011; Wilkinson & Houghton, 1982). The order of these tasks was counterbalanced in order to prevent this fact affects the results. Fifteen minutes following completion of both tasks the second saliva sample collection was performed ( $C_2$ ).

When subjects had finished all the pre-physical stress assessments, they were required to complete the stressing exercise protocol. So as to control the level of cortisol induced incremental exercise (Nicolson, 2008), fifteen minutes after finished exercise, participants carried out the third cortisol measurement ( $C_3$ ). Subsequently, the cognitive tasks were performed in the same order as in the pre-physical stress state.

Finally, to test the effect of cognitive tasks on the stress level, the fourth measurement of cortisol was sampled fifteen minutes following completion of the post physical stress cognitive tasks (C<sub>4</sub>).

### ***Fatigue exercise protocol***

Participants performed a maximal incremental protocol in treadmill (H/P Cosmos Quasar, H/P/COSMOS Sports & Medical, Nussdorf-Traunstein, Germany) to reach a physical stress state, Initial speed was 5 km·h<sup>-1</sup> with each 30 seconds speed being increased 0.5 km·h<sup>-1</sup>. The inclination was of 1 % during the entire protocol. During this protocol, subjects were monitored with a portable gas analyser (Oxycon Mobile, Jaeger, CareFusion GmbH, Hoechberg, Germany). The activity was completed when 3 out of 4 criteria were reached: i. incapacity to increase running speed, ii. respiratory exchange ratio ( $VCO_2/VO_2$ ) > 1.1, iii. theoretical maximal heart rate was reached, and iv. oxygen consumption does not increase whereas running speed increases.

### ***Psychomotor Vigilance Task (PVT)***

PVT task is based on that originally established by Wilkinson and Houghton (1982). This task was designed to measure sustained attention by recording the reaction time of participants to visual stimuli occurring at random intervals. Using E-Prime® (Schneider, Eschman & Zuccolotto, 2002) software and laptops 15" computerized stimuli to measure this function were generated. Moreover, data acquisition and data analyses were performed using this software. In each trial, black horizontal bars were shown on the screen on a grey background. Later, bars changed to a vertical position at random time intervals (2000 to 10000 ms). Participants were instructed to respond as quickly as possible to detect the change. They were required to respond with their dominant hand by pressing the laptop's spacebar. The RT was computed as the mean of time elapsed between the stimulus presentation and the response. Anticipation responses were considered as RT <150 ms. In addition, lapses (RT > 500 ms), as a key measure were analysed to examine the maintenance of surveillance Task duration was 9 minutes divided into three blocks of three minutes each. The three performance variables were obtained for each block and for the entire trial.

### ***Digit Span Task (DS)***

We used the Digit Span Tester program (GNU Public Licence, 1991), based on the subtest of the Wechsler Memory Scale III (Wechsler et al., 1997), consisting of two parts applied separately: Digits Span Forward (DSF) and Digits Span Backward (DSB). The stimulus was presented only visually. On the screen, each number of the sequences was shown during one second; with one second of pause between numbers. A blue bar with the message "Enter Response" and a text field appeared, once the sequence finished, in order to the subjects responded. The length of the sequences started at 3 and increased one by one until a maximum of 9. Two sequences with the same length of numbers were shown. The task ended when participants properly reproduced the sequence of nine digits in length, or if two erroneous sequences of the same length were introduced consecutively. DSF is considered a measure of concentration and memory. However, the DSB measures working memory, also involve executive function, by requiring active and effective processing and manipulation of information (Baddeley, 1992; Ericsson & Kintsch, 1995). The final performance indicators consist of two variables: i. accuracy (i.e., the maximum sequence length at which the two trials were correctly responded to) and ii. total score (i.e., the total number of correct sequences).

### ***Saliva sampling and cortisol analyses***

The saliva samples were collected with a "salivette" (SARSTEDT S.A., Spain). All participants received instructions for properly collecting the saliva sample. Care was taken that participants did not brush their teeth, eat or drink anything for 30 minutes before the saliva sample was taken.

Saliva samples were taken in the presence of research staff to control the study procedure and to prevent possible errors with saliva collection (such as non-compliance with instructions by participants) (Kudielka, Hellhammer, & Wüst, 2009). The waking time of the subjects was also controlled to prevent possible contamination due to the oscillation of the cortisol circadian cycle (Nicolson, 2008). Thus, research procedures and sample collection were set keeping in mind the time interval between these procedures and awakening. Samples were kept in a freezer at  $-20^{\circ}\text{C}$ , and later analysed using commercial salivary cortisol ELISA kits (DRG Instruments GmbH, Germany). All samples were analysed simultaneously and in duplicate (the mean cortisol value of this duplicate analysis was used for subsequent analysis).

### **Data analysis**

Statistical analysis was done using SPSS 20 for windows (IBM Corporation, Armonk, NY). First, we applied descriptive statistics to calculate the mean and median as measures of central tendency and standard deviation and interquartile range as measures of dispersion. Then, the assumption of normality was checked by means of (K-S test). In the case of cortisol, we applied a parametric analysis because this variable passed the normality test once log-transformation was complete. In particular, two factors repeated measures ANOVA [exercises-induced stress (pre and post physical stress) and cognition-induced stress (pre and post cognitive task)]. Follow up was carried out using pairwise comparison with Bonferroni correction.

The effects of exercise (pre and post) and time (block) on RT during PVT were tested using two factors repeated measures ANOVA. When significant effects were found, pairwise comparisons with Bonferroni correction were performed. Finally, anticipations and lapses were analysed using non-parametric test (i.e., these variables did not pass normality assumption). Wilcoxon sing rank test was specifically applied to check for differences between pre and post physical stress state. Moreover, Friedman's ANOVA was applied to determine block effects in these variables. The post hoc analysis was performed using Wilcoxon sing rank test.

As regards DS variables, Wilcoxon sign rank test was applied to establish differences between pre and post exercise. Finally, Spearman Correlations were requested to establish lineal relationships between cortisol and cognitive performance variables. Relationships between PVT and DS variables were also determined. Significance level was set at  $p = .05$  for all the analysis.

## **RESULTS**

### **Cortisol measures**

Our results show significant main effects of exercise, [ $F(1, 33) = 41.32, p < .001, \text{partial } \eta^2 = .56$ ] and cognitive task [ $F(1, 33) = 31.83, p < .001, \text{partial } \eta^2 = .49$ ], showing lower cortisol levels before the exercise and higher cortisol values before the cognitive task (figure 1).

Finally, a significant exercise  $\times$  cognitive task interaction effect was found, [ $F(1, 33) = 15.98, p < .001, \text{partial } \eta^2 = .33$ ]. As seen in figure 2, the effect of cognitive task on cortisol was only statistically significant before exercise ( $p < .001$ ).

### **Psychomotor Vigilance Task (PVT)**

There were main effects of exercise [ $F(1, 33) = 7.95, p = .008, \text{partial } \eta^2 = .19$ ] and time, [ $F(2, 66) = 19.28, p < .001, \text{partial } \eta^2 = .37$ ] on RTs. Concretely, RTs were lower before (279 ms) than after (291 ms) physical exercise. Regarding time effect, RT of block 3 was higher than in block 1 and 2 ( $p < .05$ ) while RT was higher in block 2 than in block 1 ( $p < .05$ ; figure 3). No significant exercise  $\times$  time interaction was found [ $F(2, 66) =$

2.27,  $p < .11$ , partial  $\eta^2 = .06$ ].

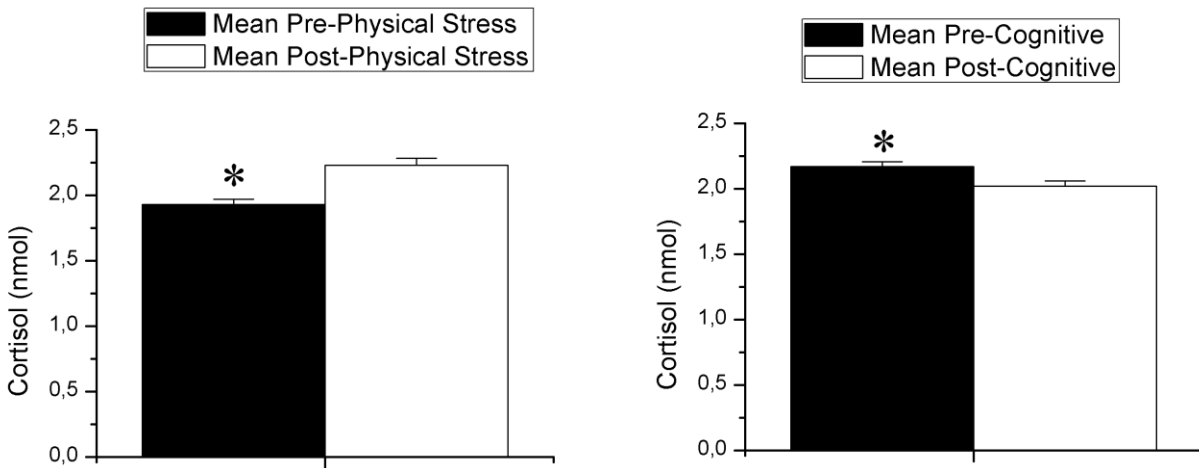


Figure 1. Cortisol data before and after exercise and cognitive tasks

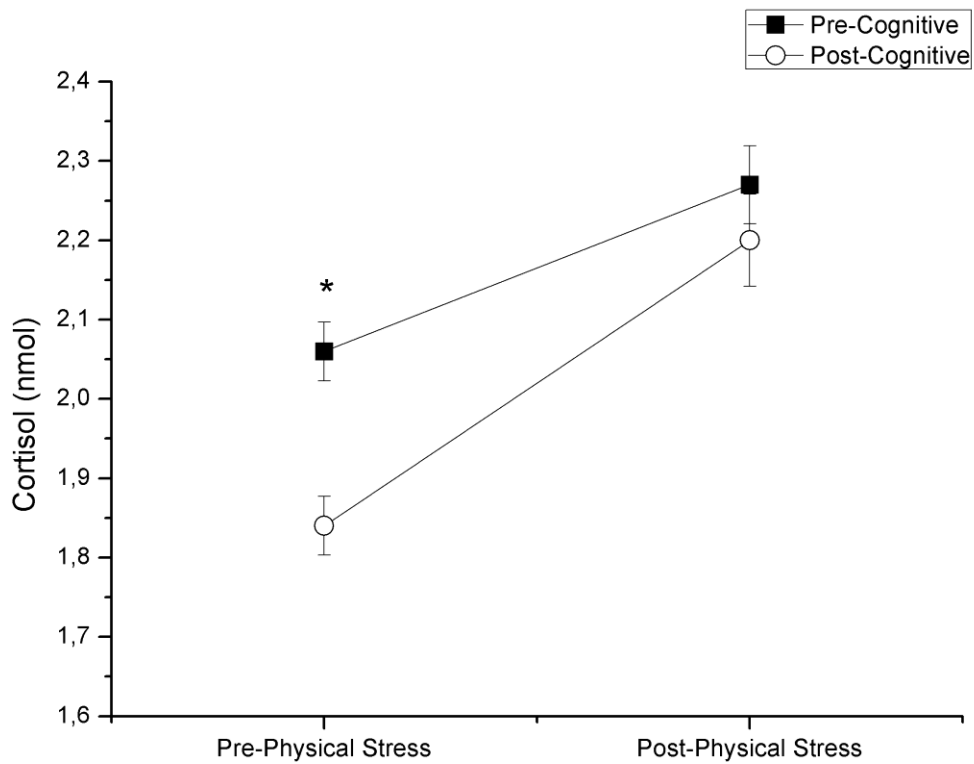


Figure 2. Interaction effect of exercise and cognitive tasks in cortisol levels

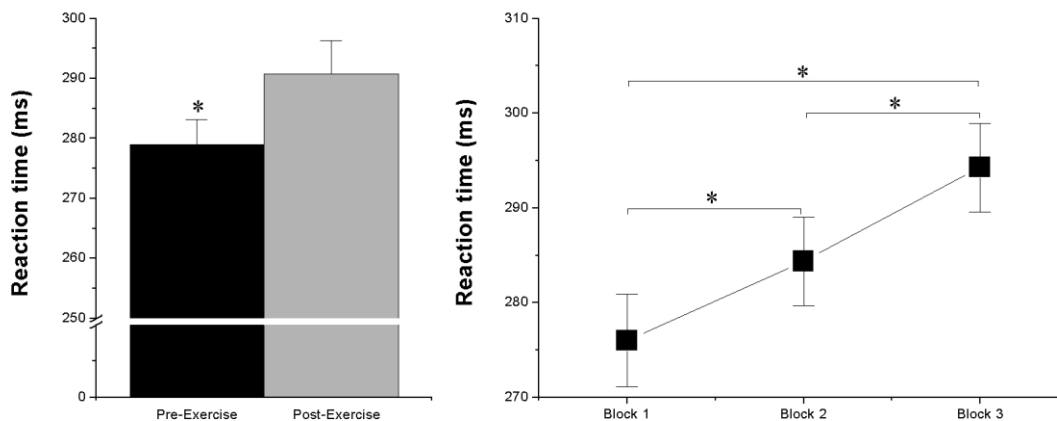


Figure 3. RT data pre vs. post-exercise and effect of time

In addition, there were significant changes in lapses due to exercise ( $z = -2.78$ ;  $p = .005$ ;  $r = -.34$ ). Lapses were lower before (Median = 2, IQR = 2) than after (Median = 3, IQR = 5.25) exercise. Nevertheless, there were no differences in anticipations between pre and post-physical exercise ( $p = .92$ ).

As for differences between blocks, there was an effect of this factor on lapses [ $\chi^2(2) = 6.34$ ;  $p = .042$ ] pre-physical stress condition. Pairwise comparisons showed lapses were lower in the second block (Median = .00, IQR = 1) than in the third block (Median = 1, IQR = 2.0,  $z = -2.06$ ;  $p = .039$ ;  $r = -.25$ ). In PVT post-physical exercise there were no such effects [ $\chi^2(2) = 3.78$ ;  $p = .15$ ]. Moreover, there was no effect of time in anticipations of pre [ $\chi^2(2) = 0.11$ ;  $p = .95$ ] and post [ $\chi^2(2) = 0.73$ ;  $p = .7$ ] exercise.

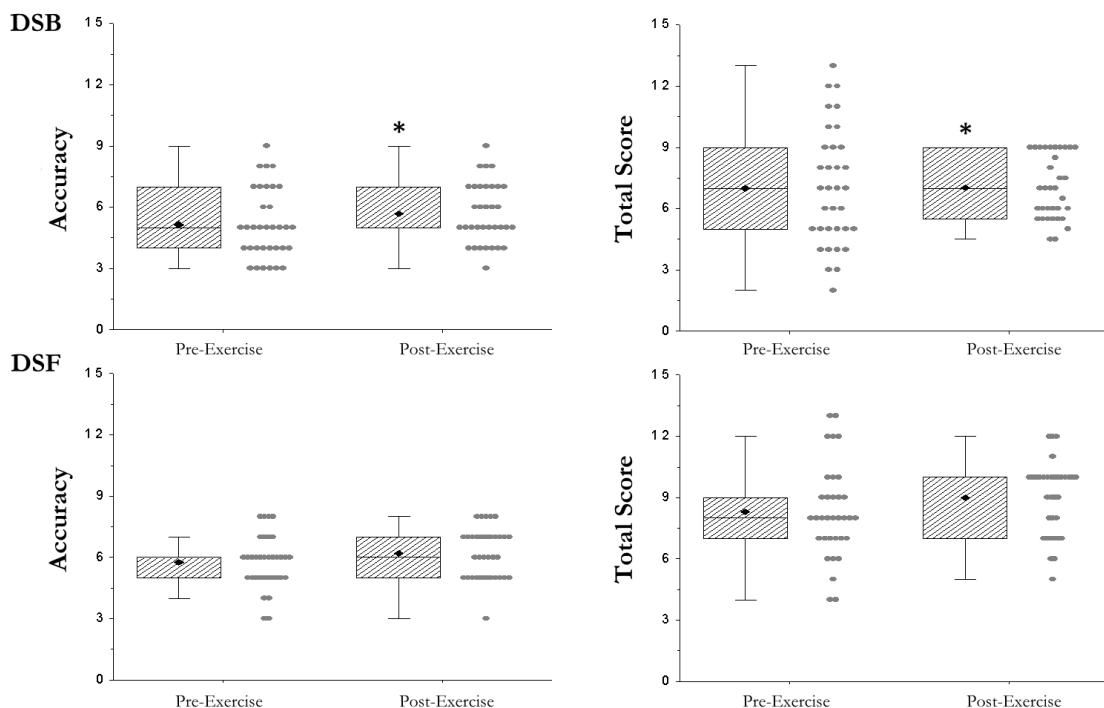


Figure 4. Digit Span variables pre vs. post-exercise



### **Digit span**

There was an effect of the exercise (pre-physical exercise vs. post-physical exercise) in accuracy [( $z = -2.10$ ;  $p = .034$ ;  $r = -.56$ )] and total score ( $z = -2.35$ ;  $p = .017$ ;  $r = -.63$ ) for the DSB. In particular, total score and accuracy were higher in post-exercise (figure 4). We found no significant differences between pre and post exercise in the variables of the DSF (Figure 4).

## **DISCUSSION**

The main aim of the present study was to evaluate how a high intensity fatiguing running protocol could affect the cortisol response and cognitive performance. The main results indicate that fatigue protocol elicited an increase in cortisol level and degrades cognitive performance in PVT (RT and lapses), improves DSB (working memory) and does not affect DSF (declarative memory). Moreover, it should be noted that the performance of the cognitive tasks reduces the cortisol level only in pre-fatigue state.

Firstly, the completion of the fatiguing protocol could lead to significant changes concerning both physiological and attentional demands. Indeed, the physiological responses to stress (i.e., cortisol) increased after the running protocol, thus confirming that such fatiguing protocols largely disturb the HPA axis (Heijnen et al., 2016; Laby et al., 2013). Regarding the cognitive task effect, there was a decrease in cortisol in the pre-exercise tests. These results seem to suggest that the cognitive tasks employed did not yield a sufficient mental strain.

In the specific case of cortisol measures after the cognitive tasks, it can be noted that the cortisol decreased in both conditions (Pre and post fatigue). Nevertheless, significant differences were only in pre-fatigue. In general, in the time course for post-stressor cortisol the maximum peak is at 0-20 min after exercise and normally the cortisol return to pre-stressor levels by 41–60 min after the end of the stressor (Dickerson & Kemeny, 2004). We must bear in mind that the acute exercise produce this fluctuation in the concentration of cortisol if the intensity of the exercise is of at least 60% of VO<sub>2</sub> max (Virtanen, 1992). Nevertheless, after high-intensity exercises (greater than 80% of VO<sub>2</sub> max) the recovery of cortisol to its basal levels can hardly change, remaining elevated for hours or even days (Nieman et al., 1994). Thus, in our study after the cognitive tasks in the post-stress condition the subjects did not achieve a significant decrease in cortisol levels (as shown in pre-stress condition) perhaps due to the high intensity of the exercise (i.e., higher than 80 % of VO<sub>2</sub> max).

With respect to PVT, RT was significantly higher in post-fatigue than in pre-fatigue conditions. The post hoc analysis revealed that this increase in RT in post-fatigue condition is not temporary or specific of one block. This fact does not coincide is contradictory with the results of other studies (Bermejo et al., 2018; Y.-K. Chang et al., 2014; Hillman et al., 2009) that suggests the transient physiological responses to exercise (e.g., endorphins or serotone) are likely to increase cognitive performance (Y. K. Chang et al., 2012). Nevertheless, the cognitive performance decreases after this transient period due to a quick cessation of these physiological responses to exercise (Y. K. Chang et al., 2012). In our case, the participant showed higher RT (i.e., lower performance) from the first block compared with the pre-exercise performance. Moreover, they continue with the normal deterioration of the performance, known as “*time-on-task*” effect (DORRIAN, Rogers, & Dinges, 2005). In addition, the accuracy of response (lapses) differs significantly between pre- and post-fatigue conditions, showing a decrease in accuracy in post-fatigue condition. Our findings coincide with previous research (Soga, Shishido, & Nagatomi, 2015) which investigated the influence of various exercise intensities on different attentional variables (i.e., RT and accuracy), confirming a deterioration after exercise of an intensity of at least 70% of maximum heart rate. In general, the impairment was characterized by significant

slower reaction times and fewer correct responses in both task conditions and particularly pronounced in the first blocks.

According to the theories of arousal, the relationship between stress and performance follows an inverted U-shaped function (Joëls et al., 2006). Thus, the time elapsed between the cessation of exercise and the evaluation of cognitive functions is another crucial variable in the psychophysiological effects of exercise induced stress (Crabbe & Dishman, 2004). This could explain worse performance in the PVT.

Moreover, we found significant differences in DSB (working memory) between pre- and post-exercise observing better results in the latter. There were no significant differences between pre and post-exercise in DSF (declarative memory). These differences could be explained because the main brain areas responsible for working memory (i.e., prefrontal cortex) and declarative memory (hippocampus) are different. In this sense, it is well known that the prefrontal cortex is more sensitive to changes in catecholamines and plasma cortisol level induced by exercise (McMorris et al., 2009; Wahl, Zinner, Achtzehn, Bloch, & Mester, 2010). Thus, improvement in working memory and not in declarative memory could be due to the higher effect of cortisol level on the prefrontal cortex area than in the hippocampus (Almela et al., 2012). This is in accordance with the dual process theory (Schneider & Shiffrin, 1977), so that working memory performance is particularly susceptible to the influence of stress when tasks are new and marginally trained. After sufficient practice, the processing becomes automatic and more effective with less demand on cognitive resources (Schoofs, Preuß, & Wolf, 2008).

This theory is supported by functional imaging studies showing that practice led to decreased activity in the prefrontal cortex while increased processing efficiency (Jansma, Ramsey, Slagter, & S. Kahn, 2001; Koch et al., 2006; Milham, Banich, Claus, & Cohen, 2003). Considering that the subjects performed the task four times it might be possible that DBS performance is particularly improved when the task processing is occupying the cognitive resources to a lesser extent.

An alternative explanation would be that adrenergic activation is essential for the impairing effects of stress-induced cortisol on working memory (Elzinga & Roelofs, 2005; Roozendaal, Okuda, Zee, & McGaugh, 2006). Considering this, during the 15 minutes of waiting (post-exercise) to collect the salivary cortisol, there is a decrease in the activation of the sympathetic nervous system (predominantly, in situations of stress and emergency) and an increase in the parasympathetic nervous system (predominates in rest).

Overall, these findings indicate that cortisol influences brain function in a time-dependent manner, affecting activity and cognitive processing brain regions in an opposite manner, in order to adapt to changing environmental demands.

## CONCLUSIONS

Following exercise-induced stress, physiological responses to stress (i.e., cortisol) increased, confirming that such fatiguing protocols largely disturb the HPA axis (Heijnen et al., 2016; Labsy et al., 2013). Additionally, the completion of the single bout of high intensity exercise did not have similar effects on the performance of the different cognitive abilities evaluated (i.e., attention, declarative memory and working memory). Indeed, exercise-induced stress had only a detrimental effect on attention without any impact on declarative memory and finding improvements in working memory performance due to his training. Koch et al., (2006) reported concordant finding by showing that the enhanced efficiency of information processing as a result of cognitive practice on working memory task. Taken together, these results underline the relative independent

effects of stress on cognitive performance seems to depend on the main brain areas responsible for cognitive functions (i.e., prefrontal cortex and hippocampus) and the time elapsed between the cessation of exercise and the evaluation of these. Hence, subjects involved in sports where cognitive abilities under physical load is important (e.g., decision-making in team sports) should benefit from specific cognitive training under stress conditions in order to minimize mistakes induced by sport and/or exercise which can alter the decision-making performance.

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