Letter to editor

Effect of Acute Sleep Hygiene on Salivary Cortisol Level Following A Late Night Soccer-Specific Training Session

Dear Editor-in-chief

We have recently shown that soccer players may benefit from acute sleep hygiene (SH) strategies to reduce the time to sleep onset after late-evening training session (Vitale et al., 2019a). In detail, soccer players allocated to an experimental group (EG, n = 17; age: 26 ± 6 years; height: 1.77 \pm 0.06 m; body mass 72 ± 6 kg) exhibited lower sleep latency (-47%; p = 0.021, ES = 0.6) and better subjective sleep quality (+11.0%, p = 0.004, ES = 0.8) respect a control group (CG, n=12; age: 25 ± 7 years; height: 1. 76 ± 5 m; body mass 75 ± 11 kg) in response to a SH protocol after a late evening small-sided-game (SSG).

During this study, we collected saliva samples (Salivette, Sarsted AG & CO, Nümbrecht, Germany) for evaluating salivary cortisol (Salivary cortisol kit, DiaMetra, DKO020) before (PRE), and after (POST), and before going to bed (POST 1) the SSG session. The cortisol awakening response (CAR) was also evaluated 30 min after morning awaking. Descriptive statistics (mean \pm SD) for the outcome measures were calculated. The normality of the distribution of salivary cortisol was checked with the Shapiro-Wilk test and two-way analysis of variance was used to test intra- and inter-group differences for salivary cortisol concentration. When a significant effect occurred, a Bonferroni's multiple comparisons test was applied. The magnitude of change after the acute sleep strategy and differences between groups were analyzed by means of a modified statistical spreadsheet, which calculates the standardized differences, or effect sizes (ES) (Hopkins et al., 2009).

Figure 1 and Table 1 present the mean $(\pm SD)$ and the raw data of salivary cortisol samples in the different time points in EG and CG. Significant intergroup variation in salivary cortisol levels in EG in PRE versus POST (p = 0.0005, ES: $\geq 2.0 \pm 0.3$, most likely), POST versus POST 1 (p = 0.036, ES: 1.4 ± 0.2 , very likely), POST 1 versus CAR (p < 0.0001, ES: $\geq 2.0 \pm 0.8$, most likely), and PRE versus CAR (p < 0.0001, ES: $\geq 2.0 \pm 0.6$, most likely). Regarding CG significant intergroup variation were observed in PRE versus POST (ES: 1.4 ± 0.7 , very likely), POST versus POST1 (p = 0.027, ES: 0.9 ± 0.3 , likely), POST1 versus CAR (p < 0.0001, ES: $\geq 2.0 \pm 0.2$, most likely), and PRE versus CAR (p < 0.0001, ES: $\geq 2.0 \pm 0.8$, most likely). Post-hoc intragroup comparisons showed that EG and CG differed in salivary cortisol concentration in CAR condition (ES: $> 2.0 \pm 0.6$, most likely). No other post-hoc intragroup comparisons in the other condition were observed. An interaction (group x time) of p=0.0013 was found.

Table 1. Mean \pm SD of the salivary cortisol levels recorded, before (PRE), after (POST), before going to bed (POST 1), and the cortisol awakening response (CAR) the evening SSG sessions.

	PRE	POST	POST1	CAR
EG	1.63 ± 0.76	3.33 ± 1.84	2.12 ± 0.72	5.13 ± 0.82
CG	$1.79{\pm}0.48$	3.37±1.42	2.04 ± 0.65	$7.00{\pm}1.07$
EC				

EG: experimental group; CG: control group.

Overall, we observed that EG and CG demonstrated similar salivary cortisol concentrations at baseline and in response to a late-night soccer-specific training session.



Figure 1. Data plot of salivary cortisol level recorded at PRE, POST, POST1 and CAR of the SSG session. Black lines denote mean values and standard deviations. EG: experimental group; CG: control group; p: p-value; n.s.: not significant; PRE: before training; POST 1: before going to bed; CAR: cortisol awaking response; **: p < 0.01 POST vs PRE EG intragroup differences; *: p < 0.05 POST1 vs PRE EG intragroup differences; ## p < 0.001 CAR vs PRE EG intragroup differences; ## p < 0.01 POST vs PRE CG intragroup differences; #: p < 0.05 POST1 vs PRE CG intragroup differences; #: p < 0.001 CAR vs PRE EG intragroup differences; ## p < 0.001 CAR integroup differences.

However, comparison of acute post intervention sleep hygiene in EG showed a reduction of CAR in EG as compared with CG. According to the objective and subjective data of our previous work (Vitale et al., 2019a) we could hypothesized that players that carried out the SH strategy before going to bed had a more restorative sleep and a reduction of the stress imposed by the training session. Our assumptions are in accordance with Rodenbeck et al. (2002) who found that athletic practice have the ability to stimulate neurotransmitters (adrenaline and noradrenaline) of the ascending arousal system, triggering the release of cortisol leading to a disruption of the sleep-wake cycle. Indeed, high levels of cortisol prior bedtime are consistent with disrupted sleep confirming an interaction between sleep and HPA axis. This is confirmed also by the recent review of Vitale et al. (2019b) who stated that impaired sleep negatively affects cortisol level with a subsequent increase of pro-inflammatory cytokines, which impedes to muscles to recover and repair from damage and increase pain perception. Moreover, according to Wright et al. (2015) if the natural circadian rhythm of cortisol is disrupted (e.g. night high intensity training), players may go into a catabolic state. This is in accordance with the recent study of Silva et al. (2019) who concluded that soccer players who exhibit poor sleep quality or nonrestorative sleep after training are associated with increased number and severity of musculoskeletal injuries.

The present study has some limitations. First, the relatively small sample size and consequent low statistical power did not allow drawing firm conclusions on the efficacy of SH to promote sleep and therefore better recovery as measured by CAR. Second, we use a short experimental design. This was use to assess the acute effects of SH. However further studies are needed to fully elucidate the long-term effect of SH on post –exercise cortisol level in soccer players.

In conclusion, present findings suggest that soccer players should be educated on the appropriate activities, behaviors, and SH recommendations following late-night Sleep hygiene and soccer

soccer practice to reduce training stress response and therefore nocturnal salivary cortisol secretion.

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