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Highlights

- A single session of anodal transcranial direct current stimulation (tDCS) is feasible to perform in elite athletes under a real world competition.
- Findings from this pilot trial presented that anodal tDCS over the dorsolateral prefrontal cortex (DLPFC) could enhance the mood state and reduce somatic and cognitive anxiety.
- In this study, competition-related output of physiological stress markers (salivary cortisol and alpha amylase) were also lower following the application of tDCS.

A feasibility study of application and potential effects of a single session transcranial Direct Current Stimulation (tDCS) on competitive anxiety, mood state, salivary levels of cortisol and alpha amylase in elite athletes under a real-world competition

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Abstract

Objective

To examine feasibility and potential effects of a single session tDCS over the dorsolateral prefrontal cortex (DLPFC) on competitive anxiety, mood state, and autonomic and endocrine stress responses in elite archer athletes under a real world competition.

Methods

Twelve male elite archers volunteered to participate in this pilot trial. Participants were randomized in order to take left anodal DLPFC, left cathodal DLPFC, or sham stimulation (the F3 or F4 areas according to the 10/20 EEG International System) in a within-subject study design. This study included three official competitions. About 45 min before the competition, the tDCS stimulation process was started and the participants were stimulated for 20 min with 2 mA current. Psychophysiological responses, including Brunel Mood Scale and Competitive State Anxiety inventory-2-Revied, were collected 15 min before each competition. Additionally, salivary cortisol (sCort) and salivary alpha-amylase (sAA) were collected 1 hour and 10 min before competition as well as 10 min and 1 hour after competition.

Results

Findings demonstrated that anodal tDCS was feasible and could lead to enhance mood state (vigor, tension and fatigue) and a decrease in competitive anxiety, as compared to cathodal and sham stimulation (all $p < 0.05$). However, self-confidence remained unaffected by the tDCS ($p > 0.05$).

Anodal stimulation resulted in a lower salivary cortisol and alpha-amylase response (all $p < 0.05$). Correlations between competitive anxiety and mood states with physiological stress markers (sCort and sAA) were not significant (all $p > 0.05$).

Conclusions

The present study provides the first preliminary evidence that anodal tDCS over the DLPFC is feasible and could modulate competitive anxiety and physiological stress responses to the acute stress of competition (potentially by a top-down regulation of HPA and SAM systems as well as the vagal system). Findings support the notion that non-invasive brain stimulation might be advantageous to enhance sport performance under competitive situations. However, additional studies in a larger sample size and different sport activities are encouraged to substantiate the findings.

Key words: Competitive anxiety, Salivary cortisol, Salivary alpha-amylase, Transcranial direct current stimulation (tDCS), Elite athlete

1. Introduction

Athletes are regularly expected to do complicated sporting skills in social evaluative and challenging environments. Competitive sports demand from athletes to perform appropriately under intense conditions, not only in the physical but also psychological context (Hagan Jr, Pollmann, & Schack, 2017). The subjective evaluation and appraisal of the athlete's ability to cope with the stressors of competition affect the development of negative emotional states and anxiety (Palazzolo, 2019). Anxiety has been explained as an emotional state, which is characterized by restlessness, worried thoughts, and a physiological arousal (Ford, Ildefonso, Jones, & Arvinen-Barrow, 2017). According to multidimensional competitive anxiety models, anxiety symptoms can be cognitive (e.g., negative thoughts, irritability, fear, feelings of weakness, and poor concentration), somatic (e.g., increase in blood pressure and heart rate, sweating, and muscle tension) and behavioral (e.g., repetitive movement and aggressive outbursts) (Carson & Collins, 2016; Martens, Vealey, & Burton, 1990; Smith, Smoll, Cumming, & Grossbard, 2006). Competitive anxiety and self-confidence are especially important in the context of sport and might be a determining factor in the final outcome of a sport competition, as it has been reported in literature (Weinberg & Gould, 2014; Craft, Magyar, Becker, & Feltz, 2003).

Prior to a stressful event (such as a world class competition) (Mellalieu, Neil, Hanton, & Fletcher, 2009) subcortical areas (such as the hypothalamus, amygdala, and brainstem monoaminergic nuclei) trigger strong (although almost unspecific) neuroendocrine responses, notably the activation of the sympathetic adrenal medullary (SAM) pathway and the hypothalamic-pituitary-adrenal (HPA) axis, leading to an increase in the concentration of cortisol (Ulrich-Lai & Herman, 2009). Cortisol is the outcome of HPA axis activation, which helps in the regulation of a stress response. In contrast to cortisol, salivary enzyme alpha-amylase (sAA) reflects activity of

the autonomic nervous system (ANS) (Engert et al., 2011; van Paridon, Timmis, Nevison, & Bristow, 2017). Some studies have already addressed elevation of salivary cortisol (sCort) and sAA levels before competition (Capranica et al., 2017; Draper et al., 2012). These responses have been correlated with sport-related anxiety (Filaire, Alix, Ferrand, & Verger, 2009; Hudson, Davison, & Robinson, 2013). Some investigations have recently indicated that increased stress biomarkers, such as sAA and sCort, before and during the competition could have adverse outcomes on athletes, eventually leading to a reduction in the overall performance of athletes (Aufegger & Wasley, 2018; Lautenbach, 2017; Lautenbach, Laborde, Achtzehn, & Raab, 2014). Therefore, interventions that can reduce psychophysiological responses of stress and anxiety are proposed to enhance athletic performance.

To test this hypothesis, researchers have investigated a diverse range of effective, safe, and noninvasive techniques with a lower incidence of side effects to improve athletic performance by reducing pre-competition anxiety (Mehrsafar & Gazerani, 2019). One of these techniques is transcranial direct current stimulation (tDCS). tDCS is a non-invasive, low-cost, and pain-free neural modulation technique that employs low-intensity direct electrical current to incite gross-defied cortical regions. This method is capable of alterations in action potentials thresholds (Knotkova, Nitsche, Bikson, & Woods, 2019). It has been implicated that the identity of these modulations is solely dependent on the stimulation polarity. It is now known that anodal stimulation is capable of increasing excitability, which is reduced in response to cathodal stimulation (Knotkova et al., 2019). A possible mechanism underlying tDCS effects might be associated with alterations in cortical neuronal activity. Although tDCS stimulates the cortical region directly under the electrode, it could also modulate subcortical structures since there are connections within the cortico-cortical neural connectivity matrices (Caumo et al., 2012).

It has been reported that tDCS could improve behavioral performance in a diverse array of cognitive domains (Coffman, Clark, & Parasuraman, 2014), balance (Pohjola, Tolmunen, Kotilainen, & Lehto, 2017), reaction time (Fridriksson, Richardson, Baker, & Rorden, 2011), motor skill acquisition (Fritsch et al., 2010), fatigue perception, and strength (Lattari et al., 2018; Reardon, 2016). These elements are considered as markers of athletic performance. Moreover, it is also thought that tDCS technique may support stress management through physiological control of the autonomic system (Montenegro et al., 2011), which could potentially result in performance gains in many sport activities. However, a recent meta-analysis has reported a weak evidence for enhancement of exercise performance by tDCS in healthy adults (Machado et al., 2019).

The dorsolateral prefrontal cortex (DLPFC) is associated with a broad spectrum of cognitive functions from emotional behavior to regulation of mood and anxiety (Cerqueira, Almeida, & Sousa, 2008). In fact, one of the essential roles of DLPFC is processing and regulating emotional responses; for example, DLPFC exhibits co-activation with the amygdala during emotion reappraisal (Davidson, 2002). The effect of tDCS on the DLPFC has been addressed in various trials across a range of patient populations and psychiatric conditions (Kekic, Boysen, Campbell, & Schmidt, 2016), with promising results. For instance, Shiozawa et al., (2014) explained the case of a treatment resistant General Anxiety Disorder (GAD) patient who underwent a three-week course of cathodal tDCS over the right DLPFC (the anode was placed over the left deltoid) who was asymptomatic both acutely and at a one-month follow-up. Another study indicated that tDCS over the DLPFC could be effective in patients with posttraumatic stress disorder (PTSD) and can improve neurophysiological and behavioral symptoms (Saunders et al., 2015). In line with this evidence, Tanaka et al., (2012) showed that the sAA levels in panic disorder increased before and after tDCS; however, cortisol did not change significantly after

stimulation. Therefore, researchers have suggested that sAA level might be a useful predictive biological marker of treatment responsiveness in patients with panic disorder. Brunoni et al., (2013) have also found a decrease in sCort levels after anodal stimulation over the DLPFC, while participants were observing emotionally negative and natural pictures. In this regard, a group of researchers reported that sCort levels decreased in anodal group and increased in cathodal group (tDCS applied over the medial PFC) after completion of the Trier Social Stress Test (TSST) (Antal et al., 2014).

Previous investigations were designed mainly to examine the impact of brain stimulation on the improvement of mental and physical performance under experimental settings (Davis, 2013). However, real world competition is typically carried out under social stressors that are capable of provoking psychological and physiological responses that can be classified into various coping strategies under competition. Although tDCS is already attracting attention for its potential ergogenic effect on athletic performance, there is limited information to confirm its efficacy, particularly in the context of real-world sports competition. Furthermore, to provide a better understanding of the actual stress of official competitions, blended assessments of psychological (e.g., mood and anxiety profile) and physiological (e.g., neuroendocrine markers) responses seem to be advantageously optimal. To our knowledge, no previous study has investigated DLPFC modulation using tDCS on physiological responses of stress, anxiety, and mood in elite athletes under real world competition. Since the DLPFC can be associated with regulation of emotional feelings, we hypothesized that application of tDCS on the DLPFC immediately prior to sports competition is feasible and might modulate the psychophysiological stress response to competition. Hence, the purpose of the present study was to explore the use and effect of tDCS on competitive anxiety, mood state, salivary cortisol, and alpha amylase levels in elite athletes. Since

investigation on the associations between emotional stress and acute physiological responses remains inconclusive in the literature, in particular within the context of official sport competition, we south to identify if an association exists between competitive anxiety and mood state with salivary stress responses to competitions under different brain stimulation paradigms.

2. Methods

2.1.Participants

Twelve male elite recurve archers volunteered to participate in this study (age 26.51 ± 2.31 years; body mass index, 23.10 ± 6.11 $kg \cdot m^{-2}$). All participants were healthy (with no psychiatric or clinical conditions), non-smokers, highly trained (archery training: 10.75 ± 2.66 $hours/week^{-1}$, aerobic training: 1.38 ± 0.34 $hours/week^{-1}$, strength training: 2.80 ± 0.41 $hours/week^{-1}$), and competed at national level. Only men were included in this study because sex-related differences in current received when the brain is stimulated transcranially has been reported (Russell, Goodman, Wang, Groshong, & Lyeth, 2014). Participants were abstained from taking stimulants (e.g., caffeine) or ergogenic aids, as well as performing intense exercise (e.g., High-intensity training) 48 hours before each competition. Participants were requested to keep their usual dietary habits throughout the study. Participants signed an informed consent form after the procedures had been explained in detail. This study was approved by the Committee of Research Ethics in Sport Sciences Research Institute of Iran and conducted according to the Helsinki declaration (Association, 2014). Participants did not receive compensation for their participation and attended voluntarily.

2.2.Design

The study was designed as a randomized, sham-controlled, within-subjects design. Participants were randomized in a counterbalanced manner to ensure all stimulation conditions (left anodal, left cathodal, or sham). To avoid carry-over effects between sessions, participants had to show up at the research center on three distinct days, with a minimal interval of 48 hours between visits (Brunoni et al., 2013). All stimulation sessions and data collection in competitions were conducted between 3 p.m. and 6.45 p.m. (see procedure). All participants underwent all study procedures, and no withdrawal or interruption occurred. A schematic representation of the experimental protocol is shown in Figure 1**.**

*****Figure 1 about here*****

2.3.Procedure

Three official archery competitions were included in this study. This enabled us to study competitive conditions similar to real-life competition. The competitions were run in accordance with the rules of the World Archery Federation. The average length of all competitions were within the range of 1 hour and 1 hour and 15 min. This study was conducted in May 2019 during the regional team selection tournament. Before the initiation of the main test, participants were familiarized with the experimental procedure in a separate session. Two saliva samples were taken on a resting day (24 hours without training), which took place 2 weeks before the competition in order to get reference values at awakening (08:00 h) and evening (20:00 h) (Figure 2 A, B). Saliva samples (2.5 ml) were obtained 60 min (15.00 h ± 30 min) before each competition. Approximately 45 min before the competition (15.15 h ± 30 min), the tDCS stimulation process was started and the participants stimulated for 20 min. Participants completed the CSAI-2 and BMS 15 min before the competition (15.45 h ±30 min), and the saliva samples were taken 10 min before competition (15.50 h ±30 min). Saliva samples were collected at 10 min (17.25 h ±30 min) and an hour after

the competition (18.15 h ± 30 min) (Filaire et al., 2009). Each athlete participated in three distinct sessions (one per each competition) in a randomized order to receive all stimulation conditions (left and/right cathodal, left cathodal/right anodal, or sham stimulation).

2.4. Stimulation

The tDCS (DC stimulator, NeuroCom, Germany) was delivered through two rubber electrodes placed in 5 cm × 5 cm saline-soaked sponges. The electrodes were placed over the F4 or F3 regions in accordance with the 10/20 EEG International System, pertaining to the regions over the right and left DLPFC, respectively. The anode was set at the F3 region and the cathode at F4 for the left anodal stimulation. Inversely, the cathode was set at the F3 area and the anode at F4 for the left cathodal stimulation. We utilized a 20-min stimulation period with 2 mA current for the actual tDCS. The apparatus was turned off after 30s of stimulation for the sham condition, as previously proven to be reliable for blinding purposes (Brunoni et al., 2013). This sham method for tDCS has no considerable neuro-modulatory effects but elicits sensations that are similar to those produced by actual tDCS (Gandiga, Hummel, & Cohen, 2006). For the assessment of blinding, we asked participates to guess whether they received sham or active stimulation when the stimulation was terminated.

2.5. Psychometric assessments

2.5.1. Competitive State Anxiety Inventory-2 Revised (CSAI-2R)

In this study, the CSAI-2R was utilized to determine competitive anxiety (Cox, Martens, & Russell, 2003). We applied the revised 17-item version from the original CSAI-2 (Martens, Burton, Vealey, Bump, & Smith, 1990), which assess the subscales *cognitive anxiety* (five items; e.g., ‘‘

I'm concerned about performing poorly''), *somatic anxiety* (seven items; e.g., ''*My body feels tight*''), and *self-confidence* (five items; e.g., ''*I'm confident I can meet the challenge*''). Answers were given on a four-point scale with the anchors (1) ''Not at all'' to (4) ''Very much so''. Higher scores on cognitive and somatic anxiety subscales denote higher levels of competitive anxiety, while higher scores on the subscale of self-confidence imply higher degrees of self-confidence. The CSAI-2R was completed 15 min before each competition.

2.5.2. Mood state

Mood state was evaluated with the Brunel Mood Scale (BMS) (Terry, Lane, & Fogarty, 2003). BMS completed along with CSAI-2R before each competition. This scale contains 24 item mood descriptors divided into six domains (*vigor, anger, depression, fatigue, tension, and confusion*) and participants demonstrate whether they are experiencing such feelings on a five-point Likert scale (4 = extremely, 3 = quite a bit , 2 = moderately, 1 = a little, 0 = not at all).

2.6. Physiological assessments

We asked our participants to refrain from eating at least one hour before saliva sampling in order to prevent contamination of saliva with drinks or food, e.g., fruit juice or coffee. We also requested participants to rinse their mouth completely with tap water before the initiation of sampling. Additionally, they were asked to not brush their teeth at least 30 minutes prior to the process of sampling. Participants were asked to passively drop their saliva in a single-use plastic cup (2.5 ml) for a 2-minute period. The cups were then transported to polypropylene vials for storage at -20 ºC until the hormonal assay. After centrifugation at 1620 × g for 15 min (to produce a clear supernatant of low viscosity), sCort levels were determined by using enzyme-linked immune

sorbent assay kits (Zellbio™, Germany). The detection limit of the kit was 0.1 nmol/L with intra- and inter-assay coefficients of variations <8%. Levels of sAA (U/ml), were determined using a kinetic reaction assay (Salimetrics, State College, PA) without any modification to the manufacturer's protocol. The intra- and inter-assay precision, expressed as percent coefficients of variations for the alpha-amylase enzyme, were less than 10%.

2.7. Statistical analyses

Data were checked for missing, outliers and normal distribution. There were no missing data in the self-reported parameters and the salivary stress markers (sAA and sCort) in any of the three stimulation conditions. The Shapiro-Wilk normality test showed most variables to be normally distributed; although, several data points were not normally distributed. We utilized LN-transformation to restored normality. However, the means in text and figures indicate absolute values for displaying purposes. The Chi-Square test was used to analyze how participants could guess if they have been in active or sham stimulation condition. To test the effect of stimulation condition on self-reported and salivary stress markers (sCort and sAA), a repeated measure analysis of variance (ANOVA) with a within-subject factor was conducted for the three conditions (sham, anodal, and cathodal). In addition, a 3 (stimulation condition: sham, anodal and cathodal) x 4 (time: -1 h, -10 min, +10 min, +1 h) repeated measures ANOVA examined competitive stress reactivity between study stimulation conditions (to determine main effect of stimulation condition, time and interaction of stimulation condition × time). Greenhouse-Geisser correction was utilized if Mauchly's Test of Sphericity was statistically significant. We applied Bonferroni correction for multiple comparisons, when the results obtained were significant. Salivary stress response analyses were also complemented by examining the Area Under the Curve (AUC) calculation with respect to the

increase (AUC_i) for competition samples. Furthermore, the relationships between psychological parameters (competitive anxiety and mood states) and salivary stress responses (AUC_i) to the competition were examined using Pearson correlation coefficients under the anodal stimulation condition.

. A statistically significant result was considered when the p-value was below 0.05. Partial eta-squared (η^2) was also reported to show the effect size (Nakagawa & Cuthill, 2007). All analyses were performed with IBM SPSS Statistics 19.

3. Result

3.1. Self-report data

No headache or discomfort was reported. Participants did not correctly guess whether they were receiving active or sham stimulation (χ^2 =3.01, $p > 0.05$). The means and standard deviations for the self-report measures (BMS and CSAI-2R) for each stimulation condition are shown in Table 1.

*****Table 1 about here*****

As shown in Table 1, the analyses yielded no significant differences in confusion (F[1.18, 13.02] = 1.130, $p > 0.05$, η^2= 0.093) , anger (F[1.30, 14.39] = 1.878, $p > 0.05$, η^2= 0.146) and depression (F[2, 22] = 2.655, $p > 0.05$, η^2= 0.194) for the stimulation condition. However, significant differences were found for the stimulation condition in vigor (F[2,22] = 6.919, $p < 0.01$, η^2= 0.386), fatigue (F[2, 22] = 6.389, $p < 0.01$, η^2= 0.367) and tension (F[2, 22] = 7.207, $p < 0.01$, η^2= 0.396) — here, *post hoc* analyses showed that vigor, fatigue and tension scores were significantly different for the left anodal vs. sham stimulation (all $p < 0.05$) and left anodal vs. left

cathodal stimulation (all $p < 0.05$). There was no significant difference between the left cathodal vs. sham stimulation (all $p > 0.05$). In fact, vigor scores were higher after left anodal stimulation, and vice versa for left cathodal and sham stimulation. On the other hand, fatigue and tension scores were lower after left anodal stimulation and higher after left cathodal and sham stimulation.

The effect on somatic anxiety and cognitive anxiety as for competitive anxiety measures, the repeated-measures ANOVAs showed significant effects for stimulation condition (F[2, 22] = 16.404, $p < 0.001$, $\eta^2 = 0.599$; F[2, 22] = 51.214, $p < 0.001$, $\eta^2 = 0.823$, respectively). In competitive anxiety subscales, pairwise comparison revealed that after left anodal stimulation somatic and cognitive anxiety were lower whereas after sham and cathodal stimulation those parameters were higher (all $p > 0.05$). Moreover, no significant differences were observed between left cathodal and sham stimulation (all $p > 0.05$). For self-confidence, however, no significant difference was obtained for the stimulation condition (F[2, 22] = 2.825, $p > 0.05$, $\eta^2 = 0.204$).

3.2. Salivary markers

Regarding the influence of tDCS on sCort, we observed a main effect of *stimulation condition* (F [2, 22] = 16.892, $P = .001$, $\eta^2 = 0.606$) and a main effect of time (F [3, 33] = 164.047, $p = 0.001$ $\eta^2 = 0.937$) as well as an interaction of *stimulation condition* × *time* (F [2.54, 28.02] = 8.992, $p = 0.001$ $\eta^2 = 0.450$). In more details for competition-related changes, no significant difference was found in sCort levels at 1 hour before competition (F [2, 22] = 3.010, $p = 0.070$, $\eta^2 = 0.215$) for stimulation condition. Figure 2 C indicates significant differences in sCort at 10 min before competition (F [2, 22] = 11.650, $p = 0.001$, $\eta^2 = 0.514$), 10 min after competition (F [1.17, 12.93] =

15.395, $p = 0.001$, $\eta^2 = 0.583$), and 1 hour after competition (F [2, 22] = 12.208, $p = 0.001$, $\eta^2 = 0.526$) for stimulation condition. *Post-hoc* comparisons showed that anodal DLPFC stimulation was the only condition associated with a significant decrease in the level of competition-related salivary cortisol. There were statistically significant decreases after anodal stimulation as compared to cathodal and sham stimulation (all $p < 0.05$). No significant difference was observed between left cathodal vs. sham stimulation (all $p > 0.05$). The subsequent analysis of AUC_i confirmed this difference between stimulation conditions (F [2, 22] = 10.232, $p = 0.001$, $\eta^2 = 0.482$; Figure 2 D). A pairwise comparison showed that the anodal DLPFC stimulation had a lower competition-related output of sCort as compared to cathodal and sham stimulation (all $p < 0.05$). Moreover, no significant difference was observed between left cathodal and sham stimulation ($p > 0.05$).

*****Figure 2 about here*****

In relation to impact of tDCS on sAA, we observed a main effect of *stimulation condition* (F [2, 22] = 9.411, $P = .001$, $\eta^2 = 0.461$) and a main effect of time (F [3, 33] = 183.462, $p = 0.001$ $\eta^2 = 0.943$) as well as an interaction of *stimulation condition* × *time* (F [2.70, 29.71] = 4.991, $p = 0.008$ $\eta^2 = 0.312$). Mirroring sCort findings, no significant difference was observed in sAA levels at 1 hour before competition (F [2, 22] = 0.683, $p = 0.515$, $\eta^2 = 0.058$); but, significant differences were obtained at 10 min before competition (F [2, 22] = 6.557, $p = 0.006$, $\eta^2 = 0.373$) and 10 min after competition (F [2, 22] = 15.929, $p = 0.001$, $\eta^2 = 0.592$) for stimulation condition (Figure 2E). *Post-hoc* comparisons showed that there were statistically significant decreases in sAA after anodal DLPFC stimulation compared to cathodal and sham stimulation (all $p < 0.05$). No significant difference was observed between left cathodal vs. sham stimulation (all $p > 0.05$). However, no significant difference was found at 1 hour after competition (F [1.20, 13.19] = 1.315, $p = 0.281$, $\eta^2 = 0.107$). This was confirmed by the subsequent comparison of AUC_i scores (F[2, 22]

= 9.055, p = 0.001 η^2 = 0.452; Figure 2F), and related *post hoc* analyses showed that the anodal DLPFC stimulation had a lower competition-related output of sAA as compared to cathodal and sham stimulation (all $p < 0.05$). Moreover, no significant difference was observed between left cathodal and sham stimulation ($p > 0.05$).

Finally, the mood states (confusion: r = 0.077, $p > 0.05$; anger: r = 0.337, $p > 0.05$; depression: r = 0.165, $p > 0.05$; vigor: r = -0.467, $p > 0.05$; fatigue: r = -0.375, $p > 0.05$; tension: r = -0.053, $p > 0.05$) and competitive anxiety subscales (somatic anxiety: r = 0.377, $p > 0.05$; cognitive anxiety: r = 0.499, $p > 0.05$; self-confidence: r = -0.103, $p > 0.05$) were not found to be significantly correlated with sCort AUC_i in anodal stimulation condition. In terms of sAA AUC_i, the analysis indicated that there was no significant correlation with BMS (confusion: r = 0.301, $p > 0.05$; anger: r = -0.186, $p > 0.05$; depression: r = -0.329, $p > 0.05$; vigor: r = 0.191, $p > 0.05$; fatigue: r = 0.419, $p > 0.05$; tension: r = 0.188, $p > 0.05$) and CASI-2R (somatic anxiety: r = 0.162, $p > 0.05$; cognitive anxiety: r = 0.081, $p > 0.05$; self-confidence: r = -0.561, $p > 0.05$) parameters under anodal stimulation condition.

4. Discussion

The purpose of this study was to determine if tDCS is feasible to perform and to identify potential effect of tDCS on psychophysiological responses of competitive anxiety and mood in elite archer athletes. Looking into results of mood state, anodal tDCS stimulation over the DLPFC could reduce tension and fatigue, and enhanced the vigor when these parameters were compared with cathodal and sham conditions. However, we did not observe any significant effects on confusion, anger, and depression, which are among the negative mood states. Recent publications have

reported similar findings relevant to mood state alterations by tDCS under clinical settings, for example in psychiatric disorders (Kekic et al., 2017; Khedr et al., 2017). However, the only significant improvement in mood induced by tDCS in male elite triathletes has been described by (Valenzuela et al., 2019), where athletes received 20 minutes of anodal stimulation of the motor cortex at 2 mA and sham tDCS. Significantly, higher vigor was found in 800-meter swimming after an actual tDCS session. This finding is similar to what we identified, increased vigor, in our study. However, a single session of tDCS could not alter mood state in healthy subjects (see for review, Plazier et al., 2012; Remue, Baeken, & De Raedt, 2016). Collectively, these findings show different outcomes depending on the tested population. This inconsistency might be due to ceiling effect, which prevents further increases in positive mood state or floor effect in negative mood state in healthy subjects with normal activation of DLPFC (Nitsche et al., 2012). In addition, application of different simulation methods among studies make it difficult for comparisons. Those differences are related to electrode placement, intermittent stimulation, devices, environmental conditions and type of sport. A standard set of guidelines in the future would help in harmonization of methodological approaches.

New ways to retain high mood levels in athletes are encouraged to be tested and employed as lower mood levels are associated with a decreased chance of success (Lane, 2015). These results could be of vast interest to athletes, as tDCS may potentially improve their mood state (especially vigor, tension, and fatigue) before competitions. Since studies in elite athletes are limited, more research is needed to determine the efficacy of tDCS on mood in elite athletes.

It has been made clear that the DLPFC is an effective site for the cortical modulation of emotional regulation (Notzon, Steinberg, Zwanzger, & Junghöfer, 2018). In terms of psychological responses to competition, our athletes receiving anodal tDCS over the DLPFC were

found with a decrease in somatic and cognitive anxiety compared with the sham and cathodal stimulation. To our knowledge, no previous study investigated the impact of DLPFC modulation using tDCS on competitive anxiety, hence comparison is not possible, as whether it is a general finding or limited to this pilot trial. However, a growing body of evidence suggests promising effects of neuro-stimulation techniques on anxiety in clinical and non-clinical setting (see for review, Kar & Sarkar, 2016). For instance, a single case study has been reported using 15 sessions of cathodal tDCS over the right DLPFC for symptoms of GAD (Shiozawa et al., 2014). The patient demonstrated a substantial reduction in anxiety. Likewise, Vergallito, Riva, Pisoni, & Lauro, (2018) showed that anodal tDCS (1.5 mA for 20 min) over the right VLPFC could reduce the negative emotions and anxiety in healthy participants. Previous studies have highlighted that modulating DLPFC activity has changed attentional bias that is relevant to the cognitive paradigm of anxiety. It is known that DLPFC anodal tDCS acutely alters the processing of threatening information (Clarke, Browning, Hammond, Notebaert, & MacLeod, 2014; Ironside, O'Shea, Cowen, & Harmer, 2016). These findings support the hypothesis that emotional regulation is processed in different neural networks and further provides a mechanism to specifically modulate anxiety. Several lines of evidence have suggested that top-down regulation of negative emotions (e.g., anxiety) are correlated with increased left DLPFC activity and decreased right DLPFC (Brunoni et al., 2013; Carnevali, Pattini, Sgoifo, & Ottaviani, 2020). This effect may contribute to the modulation of deeper cortical and subcortical structures that are involved in anxiety, such as the prefrontal cortex and the amygdala (Baeken et al., 2014). Considering small sample size of this study, and need for further investigation, a firm conclusion cannot be drawn at this point. However, we observed that a single session of anodal tDCS over the DLPFC could play a role in reduction of anxiety before competition. This might be a result of a self-regulatory process that can control

physiological arousal response. However, this theory needs further investigation to identify the mechanism underlying the observed effect.

One of the most considerable results obtained from the literature is the positive correlation between successful sporting performance and self-confidence (Hays, Thomas, Maynard, & Bawden, 2009) and the literature has documented that athletes with higher self-confidence are able to better manage their stress in competitive conditions (Hanton, Thomas, & Mellalieu, 2009). We observed that anodal tDCS over the left DLPFC (compared with sham and cathodal stimulation) did not influence self-confidence in competitive conditions. To our knowledge, ours is the first study to report on the self-confidence in athletes after tDCS (as a new technique). Further research is needed to confirm or reject this result and to determine the mechanism of tDCS on self-confidence.

Our results showed that a single session of anodal tDCS over the DLPFC compared with sham and cathodal stimulation was related to the changes in levels of physiological stress markers in official competitions. Specifically, left anodal tDCS was associated with lower sCort and sAA levels as compared to sham and cathodal stimulation. Our study has provided the first report on salivary markers of stress in elite athletes participating in a single session tDCS during a real-life competition period. However, previous models have suggested that HPA activity decreases or increases according to left and right DLPFC activity, respectively (Cerqueira et al., 2008). For instance, Brunoni et al., (2013) reported that anodal tDCS is able to reduce the levels of sCort, especially during negative imagery viewing, implying that tDCS is capable of interfering with the stress responses evoked by negative imagery in healthy individuals. Moreover, other study have indicated that the concentration of sCort is declined in the anodal stimulation (over right mPFC) and while it was increased in cathodal condition following fulfillment of the TSST in healthy

subjects (Antal et al., 2014). This may be due to differences in the participants and experimental setups in the two previous studies and our study (the presentation of emotionally negative and neutral pictures as well as psychosocial stress situation in the previous and competitive situation in the present study). Accordingly, an increase in the activity of DLPFC may affect the HPA axis through the modulation of the neural activity of subcortical structures, such as the amygdala (Liu et al., 2017). Several lines of evidence have suggested that the impact of tDCS on the HPA axis may occur by a top-down modulation. In other words, alterations in the activity of the cortex change the activity of centers associated with hormonal and sympathetic regulations, which are located in the subcortical regions and brain stem (Kelley, Gallucci, Riva, Romero Lauro, & Schmeichel, 2019). Of note, the effects of anodal tDCS on these systems occur principally in the context of the stress response (Sampaio, Fraguas, Lotufo, Benseñor, & Brunoni, 2012). Future studies could investigate whether these findings would be similar in competitive athletes.

Additionally, our result highlighted that a single session of anodal tDCS over the DLPFC, compared with sham and cathodal stimulation, was associated with a decrease in sAA levels prior to competition. Previous studies on the effects of non-invasive brain stimulation on sAA and autonomic nervous system markers are rather controversial with some studies showing decreased levels (Carnevali et al., 2020), while others report increased levels (Tanaka et al., 2012a; Tanaka et al., 2012b). In the sport context, Okano et al., (2015) reported that anodal tDCS over the temporal and insular cortex modulates the autonomic nervous system (increased heart rate variability (HRV) and decreased heart rate) during maximal exercise. Besides, Montenegro et al., (2011) utilized the anodal tDCS over the left temporal region and reported a reduction in low-frequency-HRV and an increase in high-frequency-HRV values in professional road bicycle racing. ANS responses to brain stimulation are reflected on several markers. For instance, blood

pressure, heart rate, catecholamine, body temperature, and skin response are affected. In addition, one must consider that parameters of brain stimulation may alter these responses depending on stimulation time, electrode placement, and electric current. Participants and study conditions are also influential in responses. There is no similar study in the literature that we could compare our data with in ANS responses and those studies that are available have different participants or stimulation conditions that make it impossible for a comparison. Future studies would reveal if ANS responses observed here would be reproducible under similar conditions of this study. Understanding of the mechanisms underlying the effectiveness of tDCS on a neurological level is still incomplete. It can be speculated that anodal tDCS over the DLPFC might have increased the parasympathetic modulation (vagal system) or reduced the sympathetic modulation (insula and amygdala activation), and consequently, decreased the sAA (Carnevali et al., 2020). While the present study is the first to indicate reduced sAA using tDCS in elite athletes during the competition period, future studies are required to substantiate the findings in a larger cohort and to determine the underlying mechanism(s) of this effect.

No significant correlation was found between psychological parameters and competitive physiological stress markers (sCort and sAA) under anodal stimulation condition. Investigations on the psychophysiological response to competition are to some extent contradictory (see, Ehrlenspiel & Strahler, 2012) and the relationship between psychological factors and biological markers has yet to be fully elucidated in particular when it comes to sport competition. Several studies (Chennaoui et al., 2016; Chiodo et al., 2011; Mehrsafar et al., 2019) have reported that competitive anxiety and mood states are associated with increased levels of sCort and sAA prior to competition. However, some studies (Souza et al., 2019; K. Strahler, Ehrlenspiel, Heene, & Brand, 2010) have shown that this relationship is not significant. In this vein, recent meta-analyses

(Slimani, Baker, Cheour, Taylor, & Bragazzi, 2017; van Paridon et al., 2017) have revealed that several psychosocial factors are differently associated with the emotional responses during competition period (e.g., home advantage, habituation to stressful competitions, gender, type of sport, reduced social interaction, levels of competition, warm-up, consumption of food and beverages, individual differences in stress perception, and pattern of the neuroendocrine response). In addition, a diverse range of methodological elements have been used in the current literature that makes it difficult for comparison between studies and addressing potential mechanism(s). Therefore, future studies must with larger sample size and application of similar methods could reveal if any correlation exists between emotional and biological responses under competitive situations along with brain stimulation interventions.

4.1. Strengths and Limitations

We assessed elite athletes during a competition, which allowed as to analyze the applicability and effectiveness of tDCS in a real-world sport context. The multiple stress assessments is another point of strength for this study (i.e., evaluating physiological and psychological stress markers). Furthermore, our physiological markers covered the two main stress axes, the ANS and the HPA, and saliva samples provided a non-invasive method for the analysis of both stress-related axes. However, it is worth mentioning that other stress-responsive systems (e.g., cardiovascular indices) should be investigated in future studies.

This study is not exempt from limitations. This study was performed on a small cohort to meet the exploratory nature of the study testing feasibility. Conducting research with high-performance athletes such as those assessed here (i.e., elite athletes competing at the regional level)

imposes limitations. Hence, our study must be considered as a pilot study with a small sample size, which consequently suffers from low reliability in the statistical analysis. In addition, our sample was narrow, investigating only elite male archer athletes. Future research with a larger and more representative sample size is recommended to replicate these findings. Due to time constraints before the competition, we were not able to employ thorough psychological assessments. Future studies are encouraged to broaden assessment instruments to acquire further information (e.g. reactivity to stress) related to psychological stress profiles of elite athletes. It should be noted that diurnal slopes (morning-to-evening responses) were not collected on the days of competition in our study. Thus, anticipation of the upcoming competition might have biased the morning levels. Moreover, changes in daily levels of sCort and sAA might be explained by changes in other factors, e.g., sleep parameters (Strahler, Skoluda, Kappert, & Nater, 2017). Determination of diurnal slopes of salivary markers of stress on competition days and sleep measures are also recommended.

Another limitation of the present study was the lack of measures related to brain activity or cortical excitability, which would have provided a deeper insight into the actual physiological effects of the applied tDCS protocol. Indeed, it has been shown that small changes in the tDCS protocol can result in opposite outcomes on cortical excitability (Thair, Holloway, Newport, & Smith, 2017). Considering the relatively low spatial resolution of tDCS, other brain areas besides the DLPFC could also have been modulated (Morya et al., 2019). Since adjacent brain regions are also involved in emotional processing, we cannot evaluate to what extent a possible adjacent modulation could impact the outcomes. For example, the ventromedial prefrontal cortex and orbitofrontal cortex are localized directly beneath the DLPFC and correlated with affective/emotional processing (Viviani, 2014). Additionally, future investigation might determine whether repeated sessions of stimulation would have better results or a single stimulation session,

and how long these effects would last. Moreover, studies comparing tDCS effectiveness with the efficacy of interventions through cognitive-behavioral interventions (e.g., mindfulness intervention and imagery trainings) or other brain stimulation techniques (e.g., Transcranial magnetic stimulation and Vagus nerve stimulation) are highly warranted.

5. Conclusion

Findings from this explorative study suggest that sCort and sAA— markers of autonomous and neuroendocrine systems — might change after a single-session of anodal tDCS over the DLPFC, compared with sham and cathodal stimulation, during competitive conditions. Left anodal DLPFC stimulation was also associated with lower cortisol and salivary alpha amylase levels and lower competitive anxiety, fatigue and tension as well as higher vigor. Overall, this study provided first evidence on feasibility of tDCS application for potential beneficial effects in elite athletes under competitive conditions.

Conflicts of interest

The authors report no conflict of interest.

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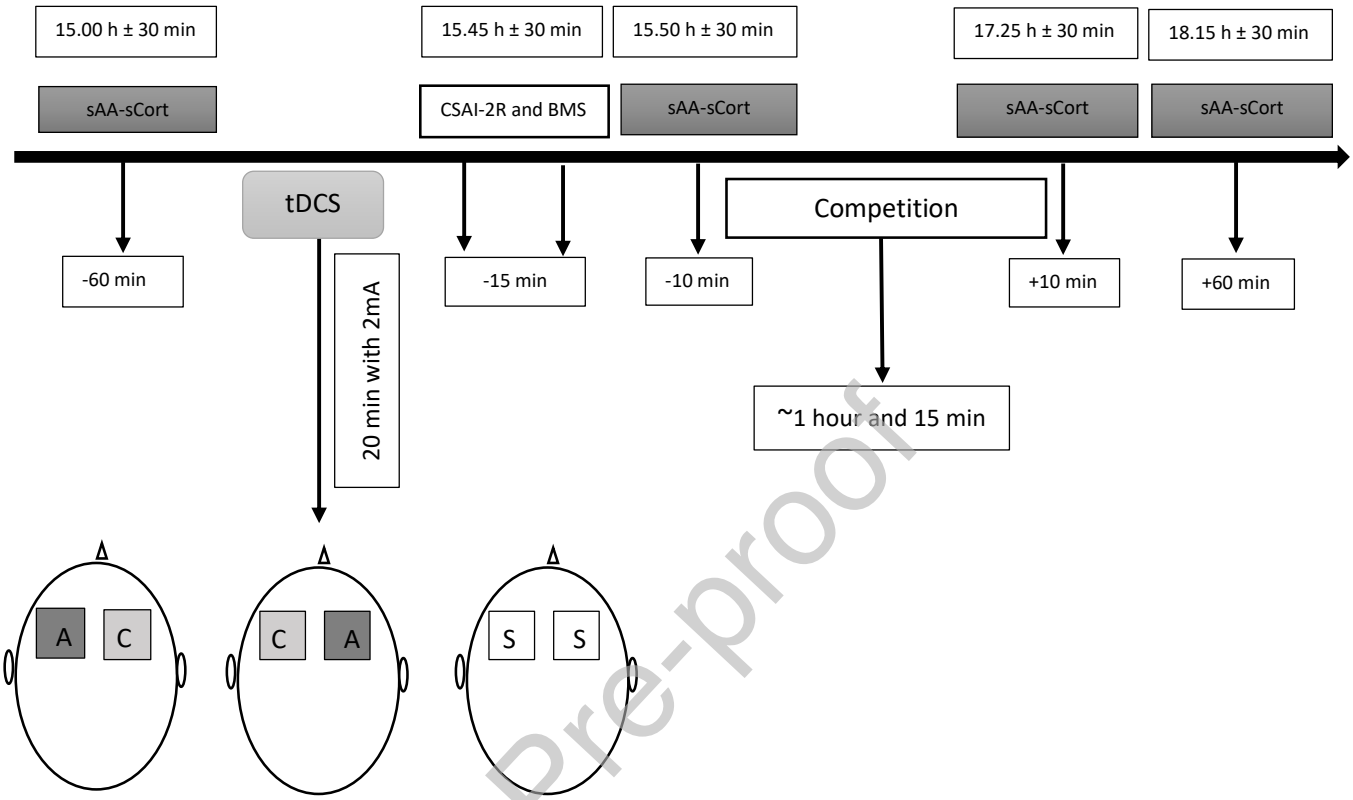
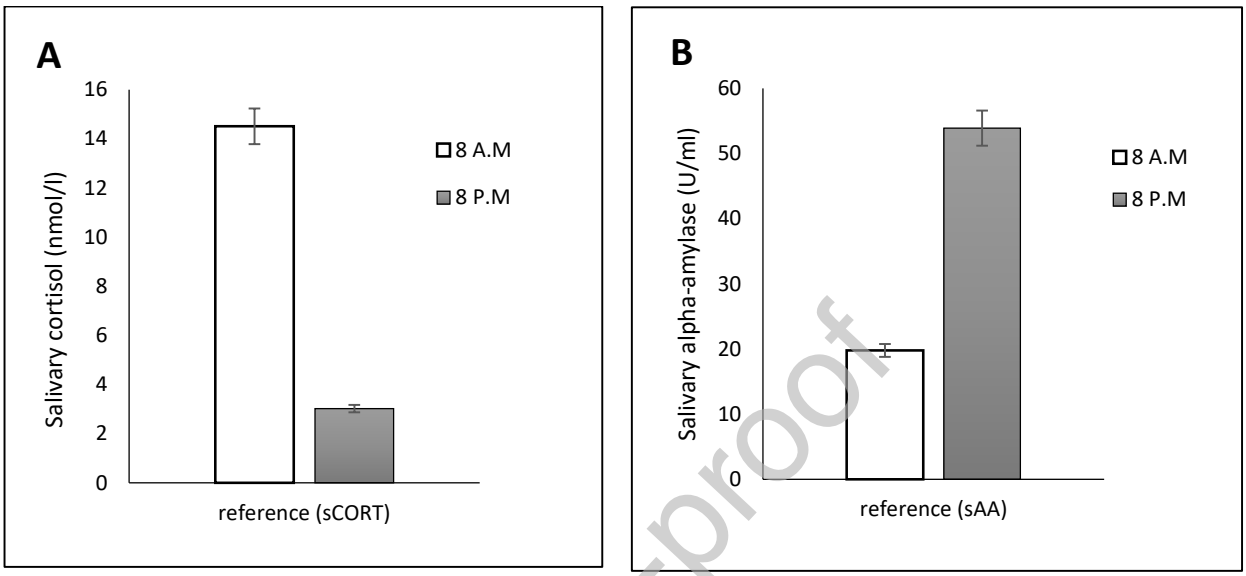
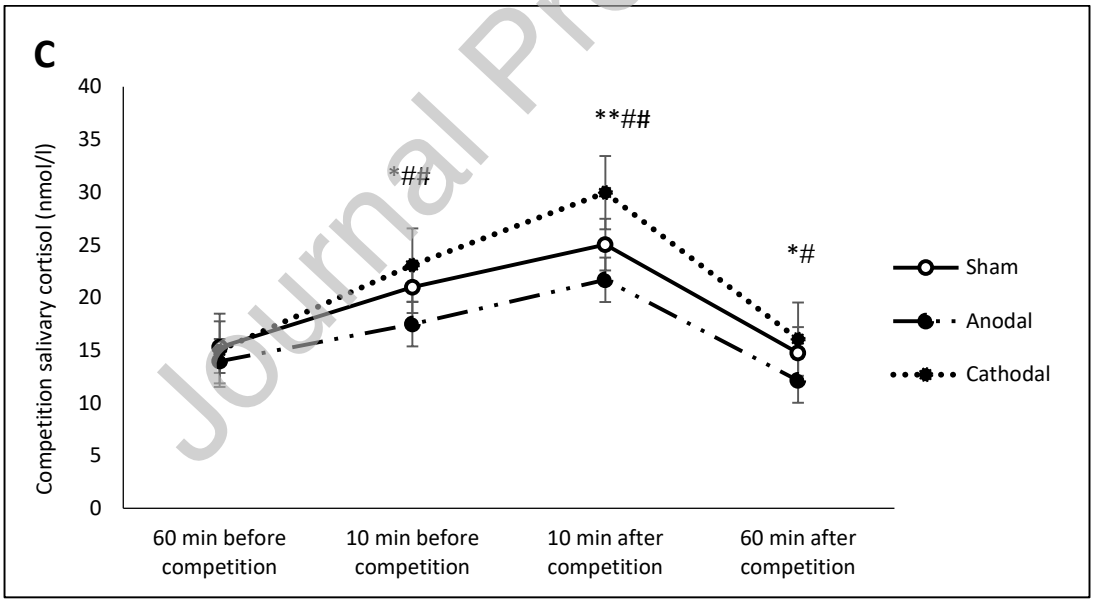
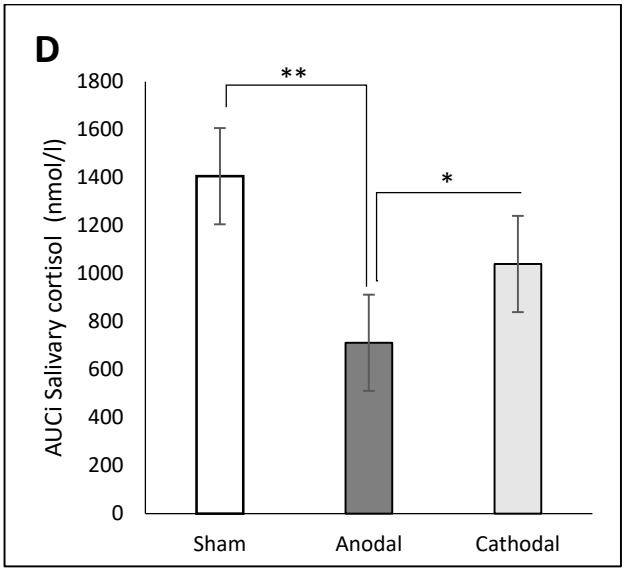


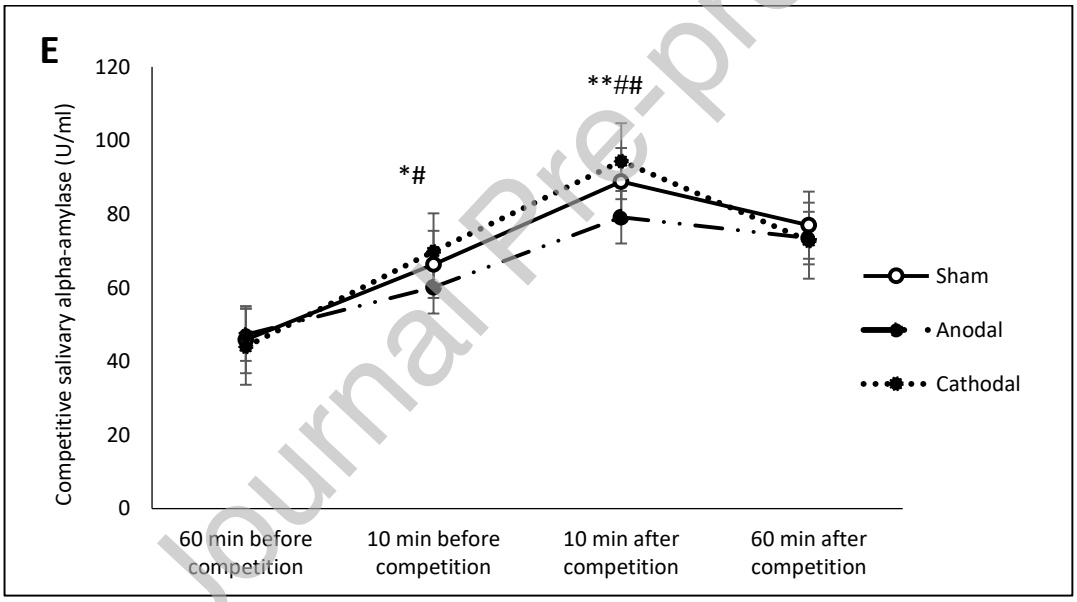
Figure 1. Schematic representation of the experimental protocol. Elite archer athletes participated in three sessions to receive all stimulation conditions (left and/right cathodal, left cathodal/right anodal, or sham stimulation over the DLPFC) for 20 min with 2 mA. Within each session, participants completed the Brunel mood state scale and competitive state anxiety inventory-2 revised 15 min before each competition. In addition, saliva samples were collected before and after each competition (1 h and 10 min before competition, 10 min and 1 h after competition). The length of all competitions were within the range of 1 hour and 1 hour and 15 min.

Note: CSAI-2, Competitive State Anxiety inventory-2; sAA, Salivary Alpha Amylase; sCort, Salivary Cortisol; tDCS, transcranial direct current stimulation. BMS, Brunel Mood Scale. A: Anodal, C: Cathodal, S: Sham.









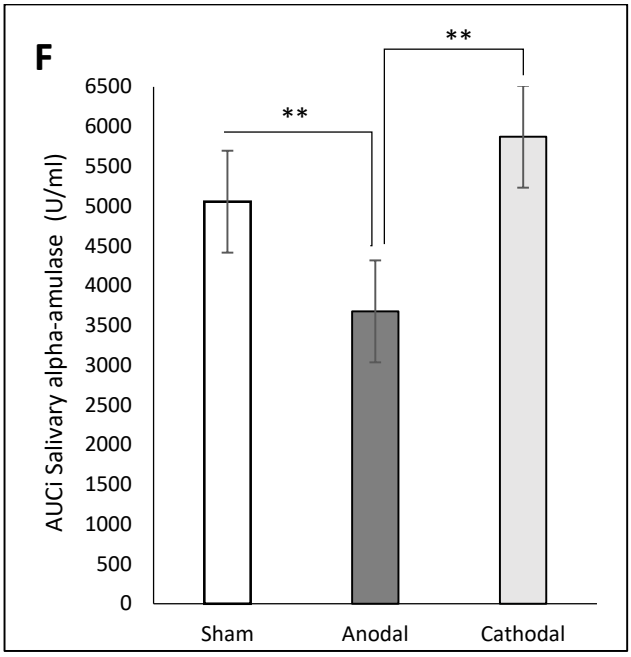


Figure 2. Morning and evening salivary samples (A: sCort, B: sAA) as reference values. These scores are shown for descriptive purposes. Competition-related changes of salivary cortisol (C, D depicts area under the curve with respect to increase [AUC_i]) and alpha-amylase (E, F depicts AUC_i) in sham, anodal and cathodal stimulation (n=12 in all condition stimulation). The length of all competitions were within the range of 1 hour and 1 hour and 15 min. In figure C and E: *$p < 0.05$ (between sham and anodal stimulation), **$p < 0.01$ (between sham and anodal stimulation), # $p < 0.05$ (between anodal and cathodal stimulation), ## $p < 0.01$ (between anodal and cathodal stimulation). In figure D and F: *$p < 0.05$ **$p < 0.01$. Each column represents the mean ratings ±S.E (standard error).

Table 1. Changes in factors of BMS and CSAI-2R for stimulation condition and results from repeated measures ANOVA

Factor	Stimulation condition			F	*p*	η^2
	Sham	Anodal	Cathodal			
Mood state scores						
Confusion	1.83 (0.71)	1.66 (0.49)	2.08 (0.66)	1.130	0.320	0.093
Anger	2.25 (0.86)	2.08 (0.79)	2.58 (0.79)	1.878	0.193	0.146
Depression	1.58 (0.51)	1.41 (0.51)	1.66 (0.49)	2.655	0.092	0.194
Vigor	9.83 (1.11)	10.66 (1.07)	9.58 (0.66)	6.919	**0.005	0.386
Fatigue	3.08 (0.79)	2.41 (0.66)	3.33 (0.88)	6.389	**0.006	0.367
Tension	2.83 (0.71)	2.08 (0.90)	3.16 (0.69)	7.207	**0.004	0.396
Competitive anxiety scores						
Somatic anxiety	12.58 (2.39)	9.41 (1.37)	12.41 (1.44)	16.404	**0.001	0.599
Cognitive anxiety	12.25 (1.96)	8.33 (1.07)	11.50 (1.16)	51.214	**0.001	0.823
Self confidence	13.33 (1.77)	14.08 (1.09)	13.16 (1.11)	2.825	0.081	0.204

Note: Each column represents the mean ± SD (standard deviation). *p<0.05; **p<0.01.