STUDY PROTOCL AND STATISTICAL ANALYSIS PLAN

TITLE: Acute responses to a potentiation warm-up protocol on sprint and change of direction in female football players: a randomized controlled study

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Introduction

A proper warm-up (WU) has the potential to enhance physical qualities, such as speed, strength and explosiveness, for a better athletic performance in training or competition (Till & Cooke, 2009). Therefore, warming up before training sessions, in addition to giving the athlete a greater physical readiness (Hammami et al., 2016; Thapa et al., 2022), is also an opportunity to mentally prepare themselves for the training contents and induce positive psychological effects, such as strengthening concentration and self-confidence (Bishop, 2003a, 2003b; McGowan et al., 2015).

In most sports, WU aims to increase muscle blood flow, heart rate, overcome viscous resistance to muscle movement, and enhance neurological excitability (Fradkin et al., 2010) in order to prepare the players for an immediate performance (McGowan et al., 2015; van den Tillaar & von Heimburg, 2016) and therefore being able to respond to physically demanding training sessions (van den Tillaar & von Heimburg, 2016; Yanci et al., 2019). To achieve this, previous research have suggested that WU protocols should have static stretching exercises (McMillian et al., 2006; Vazini Taher & Parnow, 2017) and dynamic movements that increase both body temperature and range of motion, enhance motor unit excitability, improve kinesthetic awareness and work on technique by reinforcing critical motor programs (Mann & Jones, 1999; Rutledge & Faccioni, 2001; Thompsen et al., 2007).

More recently, alternative WU protocols, focusing on potentiation methods, have been proposed in the literature (Aloui et al., 2021; Brink et al., 2022; Ciocca et al., 2021; Matusiński et al., 2021) and applied in practice. These methods are characterized by exercises that use the stretching-shortening cycle (Michailidis et al., 2019), that is, the ability of the neural and musculotendinous systems to produce maximum force in the shortest possible time (Wang & Zhang, 2016). To our knowledge, the most common WU protocol exploring the potentiating

effect presented in the literature is the one that resorts to the use of plyometrics (PLY), sprint and change of direction (COD) (Aloui et al., 2021; Michailidis et al., 2019). In potentiation, Post-Activation Potentiation (PAP) and Post-Activation Performance Enhancement (PAPE) are two of the most studied and discussed phenomena. Nevertheless, some differences distinguish these two mechanisms (Prieske et al., 2020)

PAP is associated to an increase in contractile muscle force, in response to maximal voluntary contractions and capable of conditioning subsequent muscle contractions, measured as the maximal contraction force evoked by supramaximal electrical stimulation (Blazevich & Babault, 2019; Prieske et al., 2020). From a physiological perspective, the main mechanism of PAP is the phosphorylation of myosin regulatory light chains (Blazevich & Babault, 2019; Boullosa et al., 2018; Prieske et al., 2020; Tillin & Bishop, 2009). The authors suggest that there is a time window to optimize the PAP, with a recovery period between 7 to 10 minutes (Wilson et al., 2013).

Conversely, PAPE refers to the increase in maximal voluntary strength, power, or velocity (dynamic or isometric) following a conditioning contraction (Prieske et al., 2020). Accordingly, different training protocols have been used to improve the PAPE, such as PLY (Brink et al., 2022), resistance training (Carbone et al., 2020), sprint training (Bevan et al., 2010), the flywheel paradigm (Sañudo et al., 2020) and COD drills (Lockie et al., 2014). Similarly to PAP, the performance enhancing effects may be observed after a resting period, peaking 7-10 minutes after a conditioning contraction (Cuenca-Fernández et al., 2017; Maloney et al., 2014; Tillin & Bishop, 2009; Wilson et al., 2013).

The effectiveness by which a conditioning activity can stimulate the mechanisms that strengthen muscles and improve performance depends on the balance between fatigue and potentiation (Rassier & Macintosh, 2000; Tillin & Bishop, 2009; Wilson et al., 2013). This balance is affected by a variety of factors, including training experience (Prieske et al., 2020), resting time using an optimal recovery time (Hodgson et al., 2005; Tillin & Bishop, 2009), the intensity of the conditioning activity performed (Sale, 2002), muscle fiber type and gender (DeRenne, 2010). Therefore, optimal performance occurs when fatigue has decreased, and the potentiated effect still exists (Hodgson et al., 2005). Therefore, this study aims to determine if the performance of female football players is affected after the exposure to a potentiation protocol. It is hypothesized that performance in the selected physical tests will improve significantly and meaningfully after performing the chosen potentiation protocol compared to the control group's warm-up.

Methods Trial Design

We will perform a randomized parallel trial in a single highly trained female soccer team (highly trained/national level) (McKay et al., 2022) competing in the Portuguese second division. A convenience sample will be used, due to easy access from one of the researchers, who is employed in the club. One group will receive the experimental intervention (EXPG), which will consist of a potentiation protocol as a WU and the second group will receive the control intervention (CONG) which consists of performing their usual WU exercise program. Each group will be tested before and after performing the WU to which they are assigned. Physical tests and intervention protocol will take place during the competitive period. All training sessions and physical tests will be performed on synthetic grass of the team's field. The tests will be applied without a blinded assessor integrating the technical team. However, due to the use of objective and reliable technology, hardly influenced by knowledge of the interventions to which the participants were allocated, we are confident that the non-blinded of assessors will not have a meaningful impact on the results.

Participants

Eighteen female soccer players between 16 and 21 years of age will participate in this study. Participants will be randomly allocated between a CONG (n=9) and a EXPG (n=9), using the Excel Software and the Random function and the allocation sequence will be concealed from the main researcher, responsible for the implementation of the protocols, until the beginning of the interventions. The inclusion criteria, to be applied before the randomization process, will consider athletes, from any playing position (except goalkeepers), able to train without limitations. Subjects will be excluded if they have any existing medical conditions that will compromise their participation. All participants will be informed about the purpose and content of the project, as well as the potential risks and benefits of the study, and will sign an informed consent.

Interventions

We have stipulated that the intervention will last three weeks. The first week will be a week of familiarization with the physical tests and protocol, requiring two training sessions, before the initiation of the study to accustom themselves to the physical tests and the protocol procedures. During the first week, the familiarization with the physical tests and the protocol will be performed before the training sessions. The protocol intervention will be performed in the following two weeks, in a total of four training sessions (two per week). The collection of data from the physical tests will begin in the second week, the first moment of collection will be at the at the beginning of first training session and the second moment at the end of the third week (after the fourth protocol session has been completed). The duration of each session will be 20 min.

The intervention protocol proposed in the present study was modified from the protocol used by Aloui et al. (2021), and consists of four sets: (i) First set - 6 hurdle jumps (HJ), with a

distance of 70-cm between each, followed by a 15-m sprint with COD; (ii) Second set - 6 lateral HJ (3 to the left and 3 to the right) followed by a 10-m sprint with COD; (iii) Third set - 6 bouncy strides (BC) followed by a 15-m sprint with COD; (iv) Fourth set - 6 broad jumps (BJ) followed by a 10-m sprint with COD. All COD circuits represent different configurations, changing the sprint distance and the angle of the COD curve. In the last set, we replaced the drop jump with the BJ. Due to the drop jumps being done consecutively, there is a component of horizontal resistance, so we chose to implement the BJ, which offers a greater horizontal component (Dello Iacono et al., 2017) and the forces are generated in the same musculature across the hips, knees, and ankles (Svantesson & Grimby, 1995), being a precursor exercise to improved performance in the sprint and COD (Dello Iacono et al., 2017). All repetitions and sets will be separated by 90-s recovery intervals, as well as, each set will have to be performed 3 times (Aloui et al., 2021). It should be noted that it is intended that the participants perform the procedures under study at the same time of day, i.e., same time and temperature as similar as possible. All physical tests and protocol under study are to be performed with the participants wearing their usual soccer shoes.

Outcomes

T-test

Pre-planned COD will be assessed through the T-test (Krolo et al., 2020), and applied according to standardized procedures described elsewhere (Miller et al., 2006). However, forcing the participant to touch on each cone is not uniform (Raya et al., 2013), and this step will be withdrawn. We will implement course directions used previously (Miller et al., 2006). The validated and reliable WICHRO[®] Wireless photocell system (Chronojump[®]), which consists of two pairs of barriers containing the photocell and its double reflector, will be used (López-Samanes et al., 2020), and the test will start and end on the same pair of barriers. Each player will perform each test once due to the time limitation for collecting the test data.

40-m linear sprint

The same WICHRO® Wireless photocell system will be used for the 40-linear sprint (Irurtia et al., 2022). The time will be counted in seconds and thousandths of a second (ms) with an error of \pm 0.001s (Sánchez-Pay et al., 2021). After a 5 s countdown, the participants run forward following the route marked by cones, one at 0m indicating the start place and the other at 40m indicating the finish place. Participants will start the test from a standing start position with the front foot approximately 2 cm behind the first cone.

Statistical analysis

An intention-to-treat analysis will be performed. The normal distribution will be verified by the Shapiro-Wilk test. The statistical analysis will involve descriptive and inferential statistics, and data will be presented by mean \pm standard deviation. Then, a mixed repeated measures ANOVA (i.e., within- and between-factors) will be used. The assumption of sphericity will also be analyzed. Effect sizes will be calculated using partial eta squared (η_p^2) and will be classified as small (0.010-0.059), moderate (0.060-0.137), and large (>0.137) (Cohen, 2013). Statistical significance will be assumed at p<0.05. Statistical analysis will be performed using the IBM SPSS (*Statistical Package for the Social Sciences*) version 27.

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