

Influence of Temperature on Mechanisms Inducing Post-Activation Potentiation: A Randomized Study

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Influence of Temperature on Mechanisms Inducing Post-Activation Potentiation: A Randomized Study

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“Sometimes all you need is twenty seconds of insane courage. Just literally twenty seconds of just embarrassing bravery. And I promise you, something great will come of it.”

(Benjamin Mee)

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ABSTRACT

Background: PAP is defined as an increase in the force/torque of an electrically evoked contraction, after submaximal and maximal conditioned contractions. In other words, it is an increase in peak isometric force contraction or low frequency/torque tetanic force after (i) a series of evoked stimuli, (ii) an evoked tetanic contraction, or (iii) a maximal voluntary contraction (MVC) (e.g., conditioned contraction). The study and analysis of the relationship between the physiological phenomena associated with PAP and temperature manipulation could be an asset for the targeted intervention of professionals, particularly coaches and strength and conditioning coaches.

Objectives: Identify the relevance of this phenomenon for the increase of performance in athletes of explosive disciplines of athletics and explore the relationship between temperature and the occurrence of the PAP phenomenon.

Methodology: A quantitative, cross-sectional, descriptive-correlational study with 5 athletes. The protocols consisted of 3MVC before 10 stimuli with the current defined by the analysis of the M-wave and H-reflex at the beginning of the session. Then, the cooling or heating packs (depending on the protocol) were placed around the leg for 5 minutes or the subjects remained that time at room temperature (protocol control). The protocol ended with the performance, again, of the 10 stimuli under the same conditions of current.

Results: No significant differences were found in the variables in the PAP activation exploration protocol (MVC with p=0,686; H-reflex with p=0,785; M-wave with p=0,500). Furthermore, no advantages were found in the manipulation of temperature under the mechanisms of activation of this phenomenon.

Conclusions: Further studies are needed to understand the activation of PAP mechanisms and the importance to sports performance and if cold and local heat applications genuinely modify the mechanisms of PAP activation with the exploration of different temperature manipulation methods and different exposure times to these instruments.

Keywords: Potentiation Post-Activation; Temperature; H-reflex; Maximal Voluntary Contraction; High Performance Training

RESUMO

Enquadramento: Potenciação Pós-Ativação (PAP) é definida como um aumento na força/torque de uma contração evocada eletricamente, após (i) uma série de estímulos evocados, (ii) uma contração tetânica evocada ou (iii) uma contração voluntária máxima (CVM) (ex., contração condicionada). O estudo e análise da relação entre os fenómenos fisiológicos associados à PAP e a manipulação da temperatura poderá ser uma mais-valia para a intervenção direcionada dos profissionais, nomeadamente treinadores e preparadores físicos.

Objectivos: Identificar a relevância do fenómeno PAP no aumento do rendimento desportivo em atletas de disciplinas explosivas do atletismo e explorar a relação entre a temperatura e a ocorrência deste fenómeno.

Metodologia: Estudo quantitativo, transversal, descritivo-correlacional com 5 atletas. Os protocolos consistiram em 3MVC e 10 estímulos com corrente definida após análise da onda M e reflexo H no início da sessão. Em seguida, os *packs* de arrefecimento ou aquecimento (dependendo do protocolo) foram colocados ao redor da perna durante 5 minutos, ou os indivíduos permaneceram esse tempo à temperatura ambiente (protocol de controlo). O protocolo terminou com a realização, de 10 estímulos nas mesmas condições de corrente.

Resultados: Não foram encontradas diferenças significativas nas variáveis do protocolo de exploração da ativação PAP (CVM com p=0,686; reflexo H com p=0,785; onda M com p=0,500) e não foram encontradas vantagens na manipulação da temperatura sob os mecanismos de ativação deste fenómeno.

Conclusões: Serão necessários mais estudos para entender a ativação dos mecanismos da PAP e a sua importância para a *performance* desportiva e, se aplicações de frio e calor local modificam, genuinamente, os mecanismos de ativação da PAP através da exploração de diferentes métodos de manipulação da temperatura e diferentes tempos de exposição a esses instrumentos.

Palavras-chave: Potenciação Pós-Ativação; Temperatura; reflexo H; Contração Máxima Volutária; Treino de Alto Rendimento.

ABBREVIATIONS

PAP – Post-activation potentiation

PAPE – Post-activation performance enhancement

MVC – Maximal voluntary contraction

H-reflex – Hoffman reflex

M-wave – Compound muscle action potential

M – Mean/Average

EMG – Surface electromyography

Ca^{2+} – Calcium

MU – Motor units

LABIOMEP – Porto Biomechanics Laboratory

INTRODUCTION

The neuromuscular system is extremely adaptive and complex, and different training programs, under different conditions, can change its properties (Vila-Chã, 2011). The stimulus generated by the different training programs, it undergoes changes that involve different functional and structural adaptations helping to increase the performance of the required aptitude. Based on this assumption, coaches and strength and conditioning coaches are increasingly looking for new methodologies to increase athletes' sports performance by manipulating the properties of the nervous system.

The post-activation potentiation (PAP) phenomenon, precisely defined by Sale (2002) as the increase in peak force in an isometric contraction or low-frequency force/torque after (1) a series of evoked contractions, (2) an evoked tetanic contraction, or (3) a maximal voluntary contraction (eg, conditioned contraction), is an example of one of several methodologies studied with the aim of increasing sports performance. It has been reported that, in either situation mentioned above, PAP is only effective and contributes to performance within 3 to 10 seconds after the conditioned contraction.

The occurrence of this phenomenon has been linked to the phosphorylation of myosin regulatory light chains resulting from a priori muscle contraction. These physiological mechanisms lead to increased sensitivity of actin and myosin to calcium (Ca^{2+}) and, consequently, to increased recruitment of motor units (MU) leading to enhancing the mechanical power (Force x Velocity) (Hodgson et al., 2005; Robbins, 2005, Tillin & Bishop, 2009). This change in recruitment may also be associated with increased excitability of motoneurons as a reflection of changes in the Hoffman reflex (H-reflex) (Hodgson et al., 2005).

Being a physiological phenomenon, differences in characteristics such as muscle strength, distribution of different types of fiber, training level and strength-power relationship influence the individual response to PAP (Tillin & Bishop, 2009).

In addition to the aforementioned, it is known that the individual effects of exercise are also influenced by temperature conditions. One of the conclusions

of the study carried out by Sale (2002), was that the increase in temperature would increase rate of force development and both act synergistically with PAP to improve power performance, leading, therefore, to raise the need to consider this factor in future studies. So, as the scientific evidence of the effects of temperature on PAP activation is scarce, the study and analysis of the relationship between the physiological phenomena associated with PAP and temperature manipulation could be an asset for the targeted intervention of professionals, particularly coaches and strength and conditioning coaches.

Therefore, the translation of knowledge, produced in the investigation, may contribute to the application of more enlightened and intentional training methods to athletes, simplyfying the respective adaptive processes of training and competitions, and increasing performance.

Taking into account all the information exposed so far, this study seeks to:

- Identify the relevance of this phenomenon for the increase of performance in athletes of explosive disciplines of athletics;
- Explore the relationship between temperature and the occurrence of the PAP phenomenon;

To achieve the outlined objectives, it was developed a quantitative, cross-sectional, descriptive-correlational study. Data collection was carried out at the Porto Biomechanics Laboratory (LABIOMEP).

This project is divided into four distinct chapters: i) a brief review of the literature in which the theme considered relevant is contextualized; ii) the methodology that will be followed in this investigation; iii) the results and finally, iv) the discussion, where the identified strengths and limitations of the study are pointed out, as well as the discussion, not only of the results themselves, but of their possible relationship with the investigation process.

1. LITERATURE REVIEW

This chapter addresses theoretical aspects about PAP and given the objectives of this study, a reference is made to what is known about the impact of temperature variation on athletes' muscular performance when eliciting the PAP phenomenon. Finally, a brief contextualization of the differences between PAP and, the most recent phenomenon, post-activation performance enhancement (PAPE) is conducted.

1.1. Post-Activation Potentiation

In addition to the increased body temperature, warm-up can bring other effects such as increased resting oxygen uptake, a post-activation potentiation effect influenced by the previous activity of the same muscle group, enhancing muscle contractile response and thus the ability to produce power (Silva et al., 2018). Thus, the PAP concept appears associated with warm-up in order to bring benefits in speed, strength and power in training and/or in a competitive context.

The concept of PAP, and everything related to this phenomenon and how it can be applied as a resource to increase the performance of athletes, has changed over time. In 1976, Burke et al. used this term to define the phenomenon of repetitive activation at frequencies and with numbers of pulses compatible with natural activation, distinguishing it from previous phenomena “Staircase” or “Treppe” – repeated responses of a tissue to repeated and equal stimuli increase for a time in intensity - (Bowditch, 1871) and “Post-tetanic potentiation” – increase in amplitude of twitch tension after a sustained muscle tetanic stimulation (Guttman et al., 1937).

After, Vandervoort et al. (1983) stated that skeletal muscle is overly sensitive to its contractile history, which may increase or decrease the contractile response immediately following a brief voluntary or induced contraction (5-10s), at high intensity (approximately >75% of maximal contraction), avoiding the compromise of the contractile response due to the induction of fatigue factors. Also, Sale (2002) assumes that, at a certain point, the contractile history of a muscle influences the mechanical performance of subsequent muscle

contractions, and that, in contrast to fatigue (negative effect of the contractile history of the muscle), PAP leads to performance increase. Baudry and Duchateau (2004), reinforce that the contractile response of the muscle, for the same stimulus, can be facilitated up to 200% after application of the methodology associated with PAP. A study by Gago et al. (2019), showed that PAP is a physiological process that can induce acute reductions in the delay time for force production, as well as increase it without changes in force transfer efficiency.

There are still doubts about the application of the PAP phenomenon and its role in increasing sports performance. Currently, because the different time course of effect as well as the mechanisms underpinning the force enhancement, Cuenca-Fernandez et al. (2017) proposed that the term “Post-activation performance enhancement” (PAPE) should be used when high-intensity voluntary conditioning contractions are performed to enhancing subsequent voluntary, rather than electrically evoked force production. And Prieske et al. (2020) suggested that the concept of PAP describes only the mechanistic approach, thus excluding the associated performance assumptions (described by PAPE).

In this line of thinking, PAP is defined as an increase in the force/torque of an electrically evoked contraction, after submaximal and maximal conditioned contractions. In other words, it is an increase in peak isometric force contraction or low frequency/torque tetanic force after (i) a series of evoked stimuli, (ii) an evoked tetanic contraction, or (iii) a maximal voluntary contraction (MVC) (e.g., conditioned contraction).

1.2. PAP mechanisms

Initially, according to Kilduff et al. (2007), there were only two physiological mechanisms proposed to explain this phenomenon that are still accepted today. The first is the regulation of myosin light chain phosphorylation, considered the main mechanism until nowadays (Zero & Rice, 2021), which consists of an increase in Ca^{2+} sensitivity, leading to a decrease in the distance between actin and myosin, which, in turn, will accelerate the bridging velocity. transition from

weak to stronger binding states, thus increasing the rate of force production (Macintosh, 2010).

Whereas increased sensitivity to Ca^{2+} has its greatest effect at low myoplasmatic levels of Ca^{2+}, as occurs in twitch and low-frequency tetanic contractions but, increased sensitivity to Ca^{2+} has little or no effect at saturating Ca^{2+} levels, as occurs in high-frequency tetanic contractions (Sale, 2002), so, following that thought the conditioning activity (e.g. 10s MVC) can increase due to PAP and decrease due to fatigue low- and high- frequency force, respectively.

The second is that muscle pre-contraction can increase the excitation potential along the spinal cord, resulting in increased motor unit recruitment, ie, increased alpha motor neuron excitability. An induced tetanic contraction decreases the transmitter failure during subsequent activity, via one or a combination of several possible responses wich include an increase in the quantity of neurotransmitter released, an increase in the efficacy of the neurotransmitter, or a reduction in axonal branch-point failure along the afferent neural fibre pennations (Tillin & Bishop, 2009). This adaptation leads to increased performance in the following explosive activities.

More recently, changes in muscle pennation angle have been proposed as a mechanism underlying PAP (Boullosa et al., 2018) but, as changes in muscle architecture and its role in potentiation are still not well defined, it is associated with as the least impact PAP mechanism. It is know that muscle architecture varies during dynamic and even isometric contractions; pennation angle increase (especially in transverse cross section muscles) because of training and this increase in pennation allows for a greater number of fibers to be present within a given cross-sectional area and thus is often associated with increased muscle and fascicle thickness as well as an increased strength (Mala, 2010); yet the force per cross section decrease (Ikegawa et al., 2008). In this regard, there is a greater production of force in pennated muscles, like soleus, when compared to muscles with parallel fibers (to the same volume and transverse cross section).

1.3. Factors influencing PAP

Despite the large amount of evidence in the literature, evidence for PAP effect in humans are particularly scarce. Yet, the literature suggests that the effects of PAP can be affected by several physiological and training variables, including the type of musculoskeletal fibers stimulated; the physical characteristics of the subject; the type, duration, volume and intensity of contraction and the type of subsequent activity (Gullich & Schidtbleicher, 1996; McBride et al., 2005; Sale, 2002; Seitz & Haff, 2016; Wilson et al., 2013).

Also, the conditioning of individual variability is mentioned regarding the time and degree in which the PAP phenomenon is reached. Therefore, it is a phenomenon to which some subjects respond and other do not. (Goals et al., 2016)

1.3.1. Contraction Type

The type of muscle contractions affects force-frequency relation and the range of frequencies which PAP occurs. During voluntary concentric contractions in humans, there is more myoplasmatic Ca^{2+} compared to isometric, because PAP is a Ca^{2+} dependent process may lead to a larger PAP response. Additionally, in concentric or eccentric contractions, higher frequencies are needed to evoke a given percentage of maximum force (Abbate et al., 2000) compared to isometric contractions. Yet, because fatigue and PAP can coexist (Rassier & MacIntosh, 2000) the isometric contractions are the well-explored form to produce a large amount of PAP while minimizing fatigue (Fukutani et al., 2012, 2014).

Additionally, a study made by Zero and Rice (2021) showed that for the same MVC duration, there was not significantly differences between concentric and isometric contractions. Maximal isometric contractions provide an understanding of the force producing capacity of the neuromuscular system (Hakkinen, 1994), demonstrate higher surface electromyography (EMG) activity (Hunter, 2002) and higher motor unit discharge rate (Hunter, 2003).

1.3.2. Fiber Type

Since muscles with type II fibers have a lower basal Ca^{2+} sensitivity and greater myosin light chain kinase activity than type I fibers in response to a conditioning activity, the first show greater PAP responses than type I fibers (Sale, 2002). However, it would be better for slow fibers to have greater PAP, because they are involved in low-intensity activities in which motor unit firing rates are low comparing with activities that depend on fast fibers. Despite the lower PAP capability, muscles composed of predominately type I fibers can demonstrate a PAP response (Xenofondos et al., 2015).

A practical outcome is that individuals with large portions of type II fibers develop a greater PAP benefit considering the increase in peak contraction force (Hamada et al., 2000). Consequently, PAP might improve high-speed concentric muscle force production and enhance in maximum, loaded shortening velocity (Blazevich & Babault, 2019).

1.4. Effects of PAP on training

At first, three possible adaptations on the PAP effect were suggested by Sale (2002). One would be the increased ability to activate because the magnitude of PAP is dependent on the extent of activation in conditioning activity, greater activation leads to greater PAP in the whole muscle. The second was preferential hypertrophy of fast versus slow muscle fibers commonly observed with strength training, because PAP effect is higher in type II fibers muscles. The last adaptation proposed was the increased capacity for PAP, brought about by altered myosin light chain composition.

More recently, and according to the mechanisms of PAP presented above, PAP should enhance the rate of force development during MVC as well as submaximal force for a given level of neural activation resulting in increases of force rise at specific points in jump, step, pedal, or stroke cycle when maximal force is yet to be produced (Blazevich & Babault, 2019).

Nevertheless, it´s not always clear if PAP underpins the performance enhancement, and thus peripheral and central origins have been suggested like the PAPE.

1.5. Hoffman(H) Reflex

The H-reflex, originally described by Paul Hoffmann in 1910, is an electrically induced reflex that bypasses the muscle spindle, representing a valuable tool in assessing modulation of monosynaptic reflex activity in the spinal cord. This reflex is elicited by sending an electrical stimulus along the large fast conducting Ia sensory fibers (primary afferent fibers) to the alpha motoneurons (α-MNs) in the ventral horn of the spinal cord generating an action potential toward the neuromuscular junction and produces the reflex contraction of the muscle. (Al Amer et al., 2020). The twitch response is showed in the EMG unit.

The compound muscle action potentials (M-wave) represent the summated electrical activity of the MU whose axons were depolarized by the electrical stimulus (Rodriguez-Falces & Place, 2017). The information obtained through the analysis of the M-wave is important because it allows the assessment of the peripheral properties of the neuromuscular system without direct involvement of the central nervous system and is commonly used to examine changes in sarcolemma membrane excitability (Place et al., 2010).

A percutaneous electrical stimulation of a mixed peripheral nerve, according to Tillin and Bishop (2009), causes two synchronized surface electromyography responses: the direct motor response wave (M-wave) and the reflex response (H-reflex). So, this information points to the use of the H-reflex as a way of accessing neuromuscular changes induced by different models of motor training, admitting that reflex variations occur when there are changes in the excitability of motoneurons, varying the number of neurotransmitters released or the properties intrinsic to motor neurons.

H-reflexes and M-waves are commonly assessed together by comparing the H-reflex as a fraction of M-max which allows for an estimation of the size of the motoneuron pool involved in the H-reflex. As can be seen in Figure 1, the highest amplitude M-wave (M-Max) occurs when all axons' engines are recruited, thus providing an estimate of the response given by the whole set of motoneurons (Pierrot-Deseilligny, 2000) and after a conditioned contraction, an increase in H-reflex amplitude may represent an increase in the recruitment of MU of higher dimension.

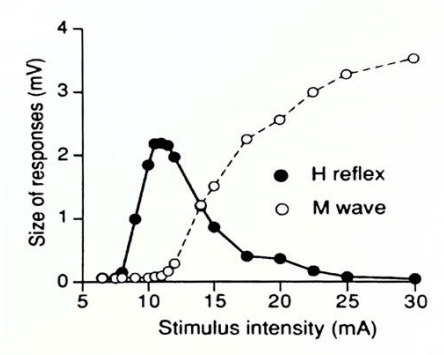


Figure 1- H and M-wave recruitment curve. The amplitudes are plotted against stimulus intensity [adapted from Pierrot-Deseilligny and Burke (2005)].

The magnitude of the H-reflex amplitude is influenced by muscle geometry (length and contraction), size of the afferent volley elicited by the stimulation pulse, background level of muscle activation, muscle contraction intensity, posture, skin resistance, different amounts of subcutaneous fat, locations of the nerve relative to the stimulus, and attention level of the subjects (Carroll et al. 2011; Klimstra and Zehr 2008; Misiaszek 2003; Palmieri et al., 2004; Zehr 2002).

The first persons to link H-reflex and PAP was Güillich and Schmidtbleicher (1996) with studies that observed a depression or enhancement of the H-reflex after a conditioning contraction.

1.6. Temperature

The effects of heat and cold strategies, measurable by performance parameters in sport, are debated in terms of periodization and impact following training (Cross, 2019) and the way our body reacts under extreme increase or decrease in temperature still raises questions.

The assessment of skin temperature has been used in several contexts to investigate different phenomena in human based on the idea that skin temperature is affected by the inflammation in underlying tissues, by the increase of metabolic activity resulted from angiogenesis, by nerve dysfunctions, or by the alteration of skin blood flow (Fernandéz, 2017), and as a non-invasive method it's used as an alternative of muscle temperature, allowing to obtain, indirectly, information of thermodynamic during exercise (Eggenberger et al., 2018; da Silva et al., 2018). However, to the best of our knowledge, no studies have used skin temperature in PAP protocols.

It is proved that the use of heat before exercise increase tissue metabolism and the muscle is easily prepared for the metabolic challenge of exercise; increases glycogen resynthesis and muscle recovery; reducing the chance of injury and energy costs (Slivka, 2012; Nape, 2010; Jarosch, 2011). Local heat modalities can be, for example, shortwave diathermy, water-circulating garments, heat wraps, whirlpool therapy, among others. (Kim et al., 2020)

Although skin temperature and muscle temperature are different, it is important to understand what changes occur when manipulating the second and how they can be similar when manipulating the first. Increases in muscle temperature are clearly and equally associated with increases in both rate of force development and shortening velocity in both fast- and slow-twich muscle/fibers (Ranatunga, 1982). It´s also affects the sensitivity to Ca^{2+}, the higher the temperature, the lower the sensitivity to Ca^{2+} (Stephenson and Willi, 1985). However, there is no evidence that these changes support the effects of PAP, other than the increase in pH (derived from the rise in temperature) which may allow PAP to be expressed (Blazevitvh & Babault, 2019).

Cooling is often used to reduce the symptoms of fatigue related to delayed onset muscle soreness, minimising muscle damage and inflammation (Bongers,2017; Ihsan, 2020). The physiological effects of interventions with cold manipulation include the reduction of muscle pain, blood flow, edema, inflammation, muscle spasms and metabolic demand. In contrast with heating applications, cooling applications are predominantly used to recover from exercise, promote return to training, and allow athletes to train at higher intensities or with greater volume and cooling modalities can be cooling vests, cold water immersion, ice slurry ingestion, cooling packs, menthol cooling, facial water spray, etc. (Bongers et al., 2017).

Despite being a topic still little explored, studies that go back to 1938 by Brown and Von Euler, showed that changes in temperature probably therefore minimally affect PAP but, as Sale (2002) refers, it is important to understand whether, practically, there are changes or not.

Back to a study by Oksa et al. (1997) to 8 subjects exposed to temperatures of 27º, 20º, 15º and 10º C (with as little clothing as possible),

showed that the subjects perform the task with better performance at higher temperatures (>= 20º), as well as the variation of each 1ºC in the temperature can modify the performance in 2-5% (depending on the time and speed of the promoted contraction). Although, Elmubarak and Ranatunga (1984) have shown that the length-tension relationship of muscle fibers (determinant in the contraction efficiency) does not seem to change at temperatures such as 27 and 15º C.

Recently, in a study by Albertas et al. (2011) was showed that during MVC, the force development variation tended to be the largest in heated muscles and the smallest in cooled muscles and warming and cooling did not affect time course of voluntary performance but affect the electrically induced muscle performance. Cooling decreased and heating increased the electrically stimulated contractile speed of the muscle compared to a neutral temperature. MVC force was reduced for muscle cooling comparing to heating and neutral. Muscle activity increased during the submaximal contractions with muscle cooling despite a lower absolute contraction force. Motor unit firing rates did not change with alterations in temperature. (Mallette et al., 2021)

Passive hyperthermia has been shown to reduce maximal muscle force as well as voluntary activation, estimated by the twitch interpolation technique (Périard, 2011). This reduction suggests that the electrical activity of the muscle during a MVC would also be reduced in hot conditions. A study by Racinais (2013), showed that the maximum voluntary torque wasn't lower, but the MVC was reduced when increased the skin temperature (25.4ºC $\pm$ 2.2 soleus temperature in normal condition to 35.2±0.9) and the EMG activity was lower compared to control conditions.

The temperature of muscles is normally below that of the core temperature (Rowell, 1974). The core temperature is protected at about 37ºC (narrow range between 33,2 and 38,2ºC) and the arms and legs tissues are kept at least 6ºC below the core, so skin temperature is typically 31ºC (Rowel, 1983). For the tissues below the skin, temperature varies by depth and location form the core, the further deep the cooler they are (Webb, 1992). In some studies, like Sawka, Cheuvront and Kenefick (2011), hot skin is defined as 35ºC and above, warm-

skin as 30-34ºC and cool/cold skin as <30ºC. In the same study, is explained that to a core temperature around 39/38ºC the skin temperature equivalent is 36ºC, so the skin temperature and core temperature gradient are fickle and alter the whole-body skin blood flow.

This information reflects the importance of studying temperature on the mechanisms associated with the activation of the PAP phenomenon.

1.7. PAPE – The “new” phenomenon

Although the objective of the study is to demonstrate the effects of the PAP phenomenon, it is extremely important to highlight the existence of a “new” phenomenon that explains in more detail the physiological processes that can determine the improvement of athletes' performance, the post-activation performance enhancement (PAPE) phenomenon.

While PAP is a physiological phenomenon whose "window of action" is short (28") and is strongly associated with the phosphorylation of active myosin chains and type II fibers, PAPE was recently proposed by Cuenca-Fernandez et al. (2017) when high-intensity voluntary conditioning contraction(s) are performed with the intent of enhancing subsequent voluntary, rather than electrically evoked, force production, whose effects can be influenced by changes in body temperature and by the accumulation of intramuscular fluid. In addition, in PAPE there is the possibility of associated neural mechanisms and, since PAPE is often undetectable at time points where PAP is maximal, some researchers have questioned whether PAP contributes to PAPE under most conditions in vivo in humans (Blazevich & Babault, 2019).

The similarities that exist between PAP and PAPE are, for example, (i) contractile force is enhanced, (ii) some delay exists before potentiation leading to fatigue, and (iii) the response is much greater in type II fibers. The differences between both are, for example, (i) the time course of their force enhancements, PAP effect within seconds and PAPE effect after minutes, (ii) PAPE is affected by muscle temperature and fluid accumulation and little is known whether PAP can be influenced by that or not, and finally (iii) the possibility exists for neural mechanisms to impact PAPE but not PAP (Blazevich & Babault, 2019).

In training practice, it’s expected that the potentiation can be achieved by execution of biomechanically similar conditioning activity at maximal or near maximal intensities. However, it is still not possible to say whether this potentiation promotes PAP or PAPE (Gepfert et al., 2020).

2.RESEARCH QUESTION AND GOALS

The research question that guided this study was:

- Is there a relationship between the decrease or increase in temperature and the PAP phenomenon?

The objectives determined for this study consisted of identifying the relevance of this phenomenon for the increase of performance in track and field athletes of explosive disciplines and explore the relationship between temperature and the occurrence of the PAP phenomenon.

3. METHODOLOGY

After reviewing the literature and defining some concepts considered relevant for a better understanding of the phenomenon under investigation, the methodological process and presentation of the material used are described.

In the development of the research, the methodological options assumed derive from the outlined objectives and, consequently, from the respective research questions to which the study seeks to answer.

This chapter describes the methodological process, starting with type of investigation, followed by the research questions, population and sample, variables, and the experimental draw as well as the temperature handling protocols, the different aspects involved in data collection (instruments, subjects' preparation, procedures, and formal and ethical considerations) and finally the statistical analysis of data.

3.1. Type of investigation

Considering the nature of the problem and in view of the objectives outlined in this investigation, a quantitative, cross-sectional, descriptive-correlational study was chosen.

The quantitative methodology focuses on the analysis of events and phenomena, as well as on the analysis of variables that can be measured, compared and/or related. As the data were collected at a time, it is cross-sectional and, since the purpose is to look for answers to describe the variables under study and to correlate, it is a descriptive study (Coutinho, 2014).

The method selected seems to be the most appropriate since it allows the collection of information, independently or jointly, on the concepts and/or variables under analysis, also offering the possibility of exploring the relationship between two or more concepts, categories, or variables to answer the research question that guide this study.

3.2. Population and sample

The number of individuals for this study were determined by calculating the sample power with G*Power software 3.1.9.7 version, having been established to reach 34 evaluated athletes for an 80% power, although it was not possible to materialize this criterion due to the exclusion criteria mentioned below and/or availability of athletes during the data collection period.

As for the inclusion and exclusion criteria, the following were defined:

- Inclusion criteria:
    - Male gender;
    - Age 18 or over;
    - Athletics practitioners in explosive disciplines;
    - At least 5 years of practice and experience in strength training.
- Exclusion criteria:
    - All individuals who do not correspond to the aforementioned items;
    - Individuals with response limitations imposed by the recent use of electrostimulation (as a therapeutic resource, e.g.) or whose H-reflex amplitude is undetectable or low due to skin resistance, locations of the nerve relative to the stimulus, among others (Palmieri et al., 2004).

Given the fulfilment of the defined criteria, the sample was constituted, successively, by all athletes, under the inclusion conditions, who voluntarily agreed to participate in the study, between June 9, 2022 and July 27, 2022.

The present study was carried out with five male federated athletes of national level, practitioners of explosive events that make up athletics such as short sprinters and horizontal jumpers. The mean age of the subjects is 21.6±4.6 years, mean height is 181.2±6.2 cm and the average weight is 72.6±6.4 kg. In addition, the subjects must have, at least, 5 years of practice in the event and 5 years of strength training experience so that the adaptations to the training are internalized.

Table 1 presents the data related to the characterization of the sample, referring to age, height, and weight.

Table 1 - Descriptive analysis of the variables under study. Age (years), Height (cm) and Weight (kg)

Subject (ID)	*Age (years)*	*Height (cm)*	*Weight (kg)*
001	29	174	72
002	23	191	78
003	20	179	80
004	18	180	68
005	18	182	65
Mean	21.6	181.2	72.6
Standard Deviation	4.6	6.2	6.4

3.3. Variables

Considering the objectives outlined, the following variables were considered for this investigation:

- Central or dependent variables:
    - Condition contraction for activation of PAP mechanisms;
    - Performance in maximal voluntary contractions.
- Independent variables:
    - Temperature for 5 minutes and a ±5ºC of difference.

The following chapter explains the experimental design, in which the operationalization of each of the variables is exposed.

3.4. Data collection: instruments

Considering the experimental design and the protocols applied during this investigation, the following instruments were used:

- National Instruments NI LabVIEW 2012 software for H-reflex and M-wave collection;

- Biodex System 4 isokinetic dynamometer;
- Electromyograph, tailor-made for investigations by LABIOMEP;
    - This is an eight channel electromyograph with preamps (AD621BNZ, Analog Devices Inc., USA) with a gain of 100 V/V and were amplified as bipolar leads with a gain of 11 V/V totalling 1100 V/V amplification. This instrument was configured for a sampling sequence of 5000 samples per second, whose protocol will consist in 3 stimuli of 5 at 5 mA, starting at 5 mA and ending at 55 mA.
- DS8R Biphasic Constant Current Stimulator from Digitimer;
- Cooling packs (cold/hot) comfort from 3M Nexcare;
    - In this study it was only used for the manipulation of cold, however the temperatures can go from -4ºC to 100ºC.
- Heat wraps by Decathlon;
    - This product allows the diffusion of a gentle heat for 6 to 10 hours, the temperature of which can reach 50ºC.
- Temperature sensor, for temperature measurement;
    - System built based on an Arduino Nano and using a 100 kohm (kilo ohm) thermistor. Data were collected at 2Hz (2 samples every second), through a serial port connection and using the PuTTY software. The sensor was previously calibrated using the Steinhart-Hart factors, obtained using the online tool available.
- Other instruments, such as blankets, blades, ethyl alcohol, among others.

Electromyographic responses to electrical stimulation of the posterior tibial nerve (M-wave and H-reflex) were recorded through a pair electrode pair of Dormo brand (30mm in diameter) with inter-electrode distance of 20mm. And the monopolar stimulation of the tibial nerve was performed through a Lifecare cathode electrode with 40x40mm (before fixing the cathode electrode, it was evaluated the position that elicited the greatest response with the least possible intensity) and a Lifecare anode electrode with 50x90mm.

The impedance and common mode rejection (CMRR) were used as recommended by Hermens et al. (1999).

3.5. Data collection: procedures

The nature of the problem and the objectives of the study determined the choice of data collection method. This was carried out in the period between June 9, 2022 and July 27, 2022, at the Biomechanics Laboratory of Porto (LABIOMEP), located in the premises of the Faculty of Sports of the University of Porto (FADEUP). For each subject, it was necessary four sessions, the first one to familiarization and the following for the application of the protocols explained below.

During any investigation process, there are ethical and moral issues that must be considered. Thus, during the entire process of this study, it was a concern to respect the formal, ethical, and legal aspects to be considered in a study with human beings and, mainly, with high performance athletes.

The research project was submitted for evaluation by the Ethics Committee of Faculty of Sport of the University of Porto. However, because this collegiate body is undergoing a change in membership, documentation has been submitted but not reviewed.

To carry out this study, an informed consent model was also given (addressed to the athletes) with mention of the intended study and its objectives (Appendix 1). In this form, voluntary participation in the study was guaranteed, and the participants were informed that the results obtained would be disseminated at scientific events. In this way, ethical principles in accordance with the Declaration of Helsinki were safeguarded.

In order to comply with the principle of self-determination, it was asserted that participation in the study was voluntary, that is, of free will, being duly informed that the withdrawal of the commitment to participation was free, at any time, without any damage associated with this decision and the confidentiality of the data collected was ensured by coding the questionnaires, replacing names with numbers and asking participants not to enter data that could lead to personal identification.

The sequence of application of the protocols was random, with a wash-out period between them to allow the carry-over effect of the previously implemented protocol not to influence the results of the following protocol. So, for the type of randomization, it was simple through a list generated by a website on the internet, and each session was at least 48 hours apart.

After consent of the individuals to participate in the study, a familiarization session was carried out so that the individual was made aware of the data collection process.

3.5.1. Subject preparation

After acclimatization of the subject to laboratory temperature, electrodes were placed in right lower limb (the limb was the same for all subjects to standardize and simplify the data collection process). The skin was lightly shaved and cleaned with alcohol and, after the skin dried, the electrodes were placed in the places recommended by Hermens et al. (1999) in the soleus of the right leg.

- Soleus:
    - Location: 2/3 of the line between the medial condylis of the femur and the medial malleolus of tibia (20mm distance between electrodes);
    - Orientation: In the direction of the line between the medial condylis to the medial malleolus.

A ground electrode was also placed on the inner malleolus of the ankle of the same leg. The monopolar stimulation of the posterior tibial nerve of the right leg was performed through a cathode electrode located in the fossa and an anode electrode placed close to the kneecap.

In addition to these instruments, the temperature sensor was also placed 1 cm away from the electrode closest to the patellofemoral joint. This sensor only served as a control to verify that the temperature reached a variation of ±5ºC during the cooling and heating of the subject.

Then, subjects were asked to sit on the isokinetic dynamometer configured as follows:

- Foot plate inclination: 0º;

- Knee flexion: 20-30º;
- Rotation axis: In the neutral position, the axis passes through the body of the astragalus, peroneal malleolus and tibial malleolus.

The right foot was attached to a specific device on the dynamometer, connected to the central tower of the equipment. The board foot support was adjusted so that the lateral malleolus was aligned with the axis of rotation of the dynamometer. Subjects kept the hip and knee flexed at 120° and the ankle at 110° of plantar flexion. All subjects were asked not to change their posture during course of the experimental protocol.

3.5.2. Familiarization session

After acclimatization to the laboratory and preparation of the subjects, in this session the subjects performed 3 MVC of the soleus muscle with a duration of 5 seconds and an interval of 2 minutes between them. During this performance, the subjects were verbally encouraged to perform the maximum force, simultaneously, visual feedback was displayed through a graphic on the computer monitor of the isokinetic dynamometer.

The highest MVC value of the soleus muscle obtained in this session was used to calculate the levels of submaximal forces used during the protocol of electrical stimulation (10% of MVC, because according to Lagerquist et al., 2006, e.g., is the value necessary to detect changes in the H-reflex). Subjects were asked to maintain a submaximal isometric contraction of 10% is the value of the force that was produced in the highest MVC and, in parallel, the electrostimulation protocol is executed. This protocol consisted of 3 stimuli of 5 at 5 mA, starting at 5 mA and ending at 55 mA, according to Vila-Chã (2011). The stimulus that will have a duration of 1ms allowing to perceive where each subject reaches the plateau in the M-wave values (balance between stimulation intensity and M-wave peak amplitude) and compare them with the values of the H-reflex. Subsequently, 15% of this amplitude value was calculated from the M wave and it was verified to which current it corresponded and if the amplitude of the reflex H was still in its ascending phase. After obtaining the necessary values, 10 stimuli were given

with the values of the current determined by the values of the M-wave and H-reflex (Control Test).

The muscle chosen was the soleus muscle, due to pre-existing studies in the area (e.g., Manal, Roberts, and Buchanan, 2008), which justify the fact that it is a muscle whose activation is uniform, compared to the gastrocnemius (e.g.). Also, Palmieri et al. (2004) explain that different levels of difficulty exist in both eliciting and interpreting H-reflexes in many muscles and soleus (or quadriceps) H-reflexes are the most widely assessed and reported.

The following chapter explains in detail the protocols used in data collection, which describes how the temperature was manipulated to achieve the objectives defined in each protocol.

3.5.3. Protocol application sessions

In the sessions dedicated to data collection, the same order of evaluation of the various parameters referred to in the familiarization session were made, apart from the electrostimulation protocol. The subjects performed 3 MVC, followed by a 2-minute period of isometric strength maintenance in the value of 10% of the highest MVC obtained in the familiarization session and then, was applied the Control Test. The assignment of the protocol to each session was done randomly, as already mentioned, to remove the dependence of the performed sequence from the analysis of results.

In the cold session (Protocol A), after the process described above, the cooling packs were applied around the right lower limb for 5 minutes (the subject remained in the same position). After this time, and a temperature change (±5ºC initial temperature difference) was verified by the temperature sensor, the subject performed 3 MVC and the Control Test.

In the neutral session (Protocol B), after the process described above, the subject remained for 5 minutes at room temperature. Then, performed 3 MVC and the Control Test.

In the hot session (Protocol C), after the process described above, the heat wraps were applied around the right lower limb for 5 minutes with blankets to lead to minimal heat loss (the subject remained in the same position). After this time,

and a temperature change (±5ºC initial temperature difference) was verified by the temperature sensor, the subject performed 3 MVC and the Control Test.

3.6. Experimental design

The experimental design (Figure 2) is materialized, whose methodological options for this empirical investigation derive from the objectives outlined.

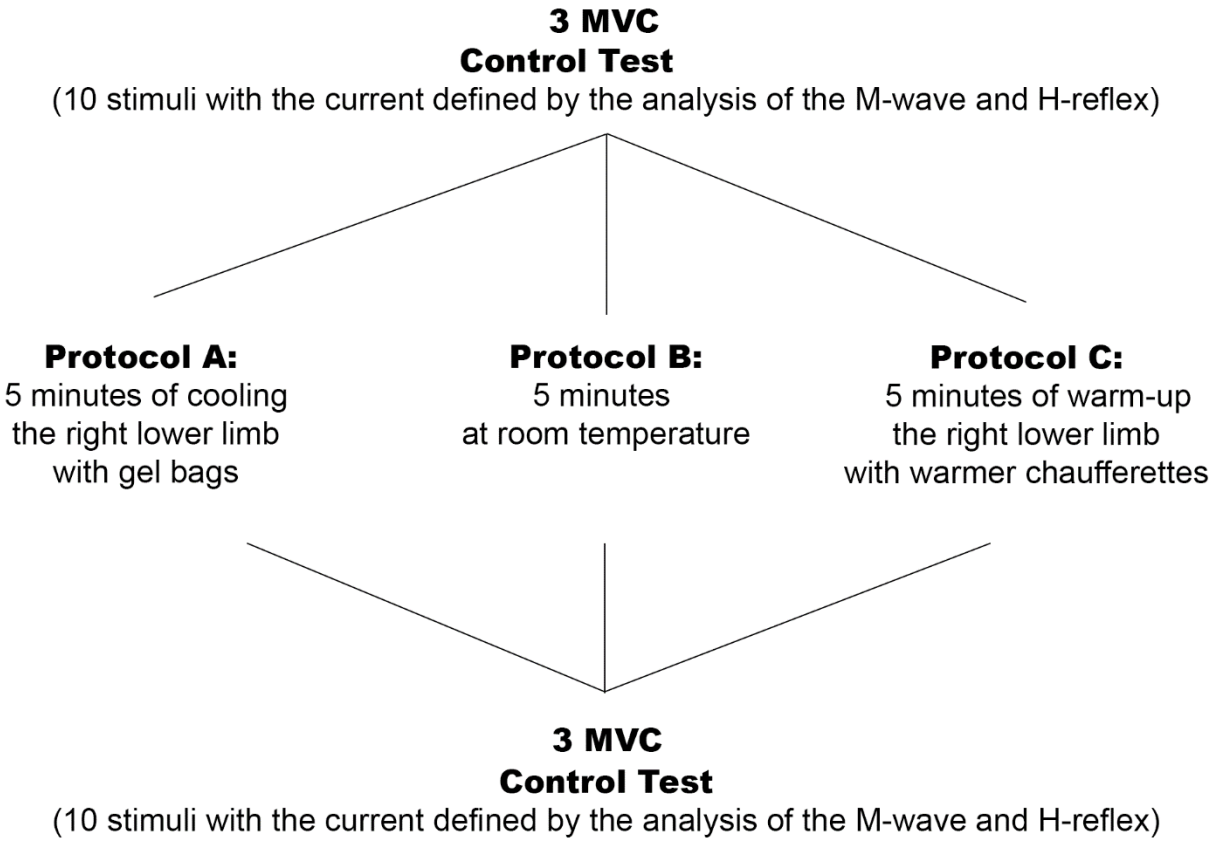


Figure 2-Experimental design of the protocols implemented.

3.7. Temperature handling protocols

There is no literature that associates the PAP phenomenon with the manipulation of lower limb temperature in the performance of a stimulation protocol, which led the researcher and research team to reflect on the most appropriate method to obtain the most reliable and valid results.

Temperature manipulation protocols will then be explained in detail in the chapters that follow.

Protocol A - 3MVC before 10 stimuli with the current defined by the analysis of the M-wave and H-reflex at the beginning of the session were performed. Then, the cooling packs were placed around the leg for 5 minutes and the temperature change data during this time was taken through the previously placed sensor. The protocol ended with the performance, again, of the 10 stimuli under the same conditions of current.

Protocol B - 3MVC before 10 stimuli with the current defined by the analysis of the M-wave and H-reflex at the beginning of the session were performed. Then, 5 minutes at room temperature. The protocol ended with the performance, again, of the 10 stimuli under the same conditions of current.

Protocol C – 3MVC before 10 stimuli with the current defined by the analysis of the M-wave and H-reflex at the beginning of the session were performed. Then, the heat wraps were placed around the leg for 5 minutes (using blankets to prevent heat dissipation over time) and the temperature change data during this time was taken through the previously placed sensor. The protocol ended with the performance, again, of the 10 stimuli under the same conditions of current.

3.8. Data processing

LabView was used for data processing, the peak of the H-reflex was normalized to the peak to peak of the M-wave and, eventually, this value was filtered between 20 and 40 Hz.

3.9. Statistical analysis of data

Regarding the analysis of the quantitative data collected, initially, the documents obtained by the different software were transcribed to an Excel sheet and, later, analysed in the computer program IBM SPSSS, version 27.

To systematize the information provided by the data, descriptive statistics techniques were used in which measures of central tendency (mean and median) and variability (standard, minimum and maximum deviation) were determined. Wilcoxon non-parametric test (chosen due to the small sample size) was also used, to determine whether there are differences (i) in the variables under study between the first and second moments, (ii) in the two moments in each protocol, and (iii) between the protocols in the second moment (after application of the protocols).

Considering the type of investigation implicit in this study, a quantitative, cross-sectional, descriptive-correlational study, and the number of subjects under analysis ($N<30$), the nonparametric Wilcoxon signed-rank test was used to analyse the results to achieve the proposed objectives and answer the research question.

For all statistical procedures, a significance level of $p<0.05$ was assumed, and for effect size <0.30 (small effect), $0.30-<0.50$ (moderate effect) and ≥ 0.50 (large effect). The results of the statistical analysis developed are presented in the form of tables and graphs, in the following chapter.

4. RESULTS

During this chapter, the results related to the variables under study (MVC, normalized H-reflex and M-wave mean) are presented and analysed to improve knowledge about the mechanisms of PAP activation, based on the outlined objectives.

For a better understanding of the results, an analysis from the general to the particular is carried out, considering “p” as the indicator of statistical significance for values <0,05 and “r” the effect size to understand how meaningful the relationship between variables or the difference between groups.

4.1. Analysis of MVC, H-reflex and M-wave peak to peak values between the protocols at the two moments

In Table 2, where it is tested whether there are significant differences between the two moments for each variable under study, no statistically significant differences were identified in the values. For the values of MVC, the subjects did not improve the performance between moment 1 (M=153.29) and moment 2 (M=147.32), z= -1.704, p=0.088, r= -0.311. In H-reflex, the values also do not present significant differences between moment 1 (M=0.23) and moment 2 (M=0.23), z= -0.505, p=0.614, r= -0.092, and finally, the results of the M-wave do not present significant differences between moment 1 (M=199) and 2 (M=277), z= -1.036, p=0.300, r= -0.189. Considering the r value associated with each variable in this analysis, it is possible to say that we are facing a small effect size value.

Table 2-Differences of variables between the first and second moments

	Sig.
MVC 1st Moment vs MVC 2nd Moment	0.088
H_Reflex 1st Moment vs H-reflex 2nd Moment	0.614
M_Wave 1st Moment vs M-wave 2nd Moment	0.300

Note: MVC=Maximal Voluntary Contraction; Sig.=Statistical significance

4.2. Analysis of MVC, H-reflex and M-wave values in each protocol at the two moments

Considering that no statistically significant differences were found between the moments (before and after application of the protocols), it is necessary to analyse the data in each variable, protocol, and moment. The MVC values, normalized H-reflex and M-wave averages obtained at the two moments in each protocol are represented in the 3rd graph. In all protocols, there was a decrease in MVC values from the first to the second moment, this change being more evident in protocol A with a 10% of decrease between moments. In normalized H-reflex values it is verified that, except for protocol A, the values obtained from the first to the second moment are the same. In protocol A there is a difference of 4% between the moments. Watching the graph 3 and respective table (table 3), the M-wave values in protocol B did not undergo major changes between the first and second moments, in protocol A and in protocol C there is an increase of about 49% and 68% respectively.

For a better understanding of these data mentioned above, table 3 shows the differences between the two moments studied in each protocol considering the variables, with no significant differences to be mentioned, except for the MVC values in protocol A (M=153.23, z= -2.023, p=0.043 and r= -0.640). In the variable MVC and H-reflex, it is possible to observe that the significance values of protocol B and C are similar (z= -0.405, p=0.686, r= -0.128; z= -0.272, p=0.785, r= -0.086 and z= -2.271, p=0.786, r= -0.718), as well as in the variable M-wave between protocol A (M=229.5) and B (M=1.97), z= -0.674, p=0.500, r= -0.213. For the values obtained, two comparisons were found with moderate effect size (MVC in protocol A and H-reflex in protocol C) showing a moderate impact of the protocols in both variables, all other have a small effect size.

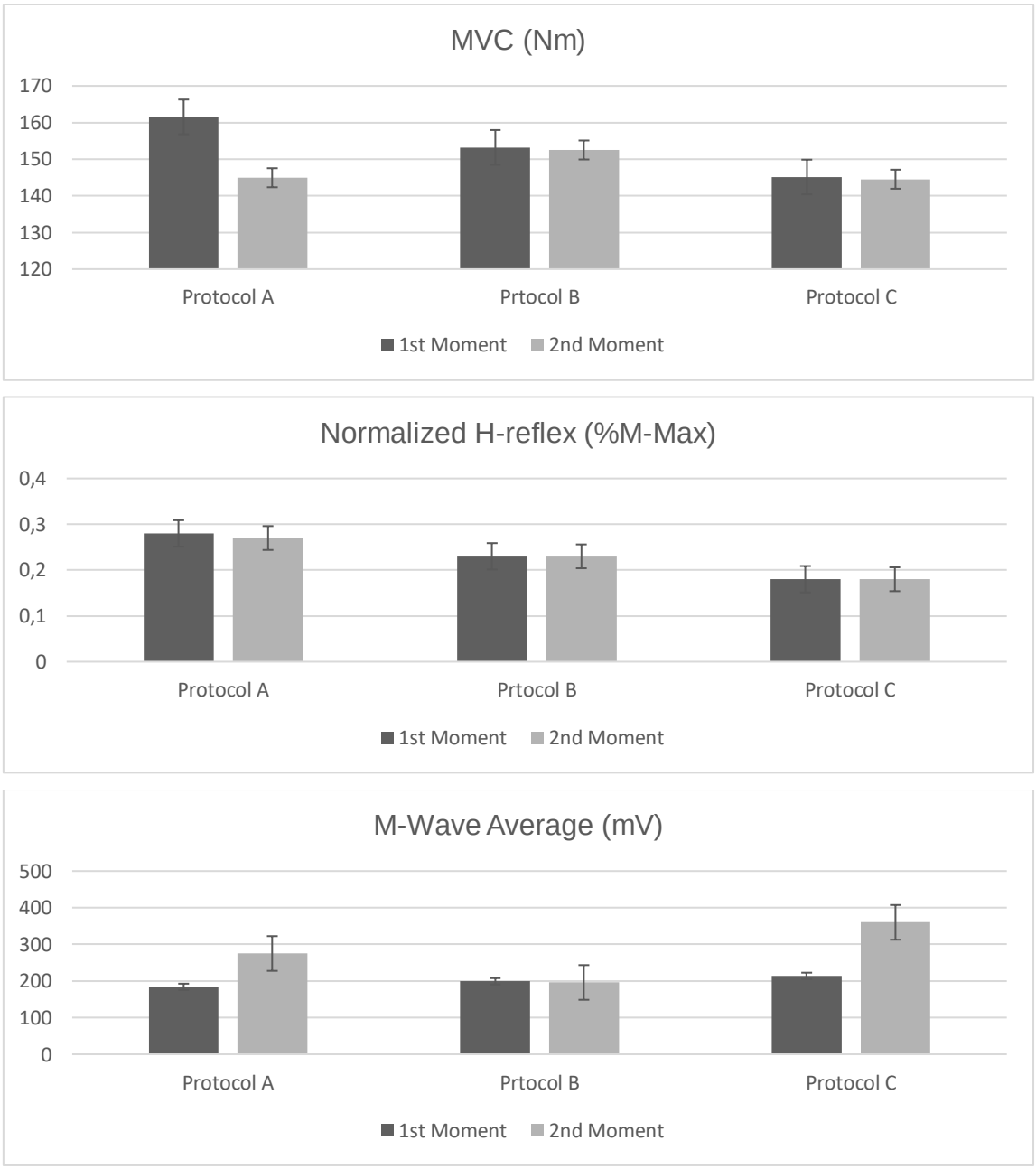


Figure 3-Graphs of MVC average values, normalized H-reflex and M-wave average at the two moments in each protocol

Table 3-Difference between the two moments studied in each protocol considering the variables

	MVC	H_Reflex	M_Wave
Protocol A	0.043	0.715	0.500
Protocol B	0.686	0.785	0.500
Protocol C	0.686	0.786	0.175

Note: MVC=Maximal Voluntary Contraction

4.3. Comparation between the variables in the first and second moments separately

To test whether the sample was in the same conditions at the beginning of each session, the differences between the variables in the first moment (before applying the protocol) were compared in table 4 and, there is no significant differences between the protocols in any of the variables. For the values of statistical significance p=0.345, z= -0.944, r= -0.298; for p=0.343, z= -0.948, r= -0.300; p=0.279, z= -1.084, r= -0.343; and, for p=0.500, z= -0.674, r= -0.213. In this table, before the compared values, all the relations present a small effect size.

Table 4-Comparation between the variables under studied in the first moment

	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**
1.MVC Pre A	-	0.345	0.345	-	-	-	-	-	-
2.MVC Pre B	-	-	0.345	-	-	-	-	-	-
3.MVC Pre C	-	-	-	-	-	-	-	-	-
4.H-R Pre A	-	-	-	-	0.343	0.345	-	-	-
5.H-R Pre B	-	-	-	-	-	0.279	-	-	-
6.H-R Pre C	-	-	-	-	-	-	-	-	-
7.M-W Pre A	-	-	-	-	-	-	-	0.345	0.345
8.M-W Pre B	-	-	-	-	-	-	-	-	0.500
9.M-W Pre C	-	-	-	-	-	-	-	-	-

In table 5, which correlates the variables under study in the different protocols only for the second evaluated moment (after protocols application), there is no significant differences between the protocols in any of the variables. The protocols that present values whose statistical significance is closer to 0.05 are in the MVC variable and in the H-reflex between protocol A and B (z= -0.674, p=0.500, r= -0.213; z= -1.483, p=0.138, r= -0.469), and in the M-wave between protocol B and C (z= -1.483, p=0.138, r= -0.469). In the values mentioned above, the effect size was tested, showing a limitation of practical applications (small effect size).

Table 5-Comparation between the variables under studied in the second moment

	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**
1.MVC Post A	-	0.500	0.893	-	-	-	-	-	-
2.MVC Post B	-	-	0.686	-	-	-	-	-	-
3.MVC Post C	-	-	-	-	-	-	-	-	-
4.H-R Post A	-	-	-	-	0.279	0.138	-	-	-
5.H-R Post B	-	-	-	-	-	0.144	-	-	-
6.H-R Post C	-	-	-	-	-	-	-	-	-
7.M-W Post A	-	-	-	-	-	-	-	0.686	0.500
8.M-W Post B	-	-	-	-	-	-	-	-	0.138
9.M-W Post C	-	-	-	-	-	-	-	-	-

Note: MVC=Maximal Voluntary Contraction; H-R= H-reflex; M-W= M-wave; Post A/B/C= Post Protocol A/B/C

5. DISCUSSION

In this chapter, a detailed interpretation and discussion of the main results obtained is carried out, seeking to highlight the resulting implications for practice in high-performance training.

5.1. Findings

The aim of this study was to identify the relevance of this phenomenon for the increase of performance in athletes of explosive disciplines of athletics and explore the relationship between temperature and the occurrence of the PAP phenomenon. To achieve these objectives, we tried to analyze the effects of three different protocols on the activation of PAP mechanisms, comparing the performance of subjects in MVC, variations of the H-reflex and M-wave.

5.1.1. Post-activation potentiation effects in athletes' performance

The manipulation of acute and chronic factors associated with sport are exploited by coaches and athletes to increase sports performance. Post-activation potentiation is one of many concepts that exist and shown promise in healthy, trained athletes to help improve acute increases in strength and power (Lorenz, 2011). However, there are still few studies whose methodology meets the most recent definition of the PAP phenomenon, which makes it difficult to understand whether the activation of this mechanism actually leads to changes in the performance of athletes.

PAP is recently defined as an increase in the force/torque of an electrically evoked contraction, after submaximal and maximal conditioned contractions. In other words, it is an increase in peak isometric force contraction or low frequency/torque tetanic force after (i) a series of evoked stimuli, (ii) an evoked tetanic contraction, or (iii) a maximal voluntary contraction (MVC) (e.g., conditioned contraction). There are many studies that seek to understand the PAP and performance link, but these studies do not consider the PAP and PAPE distinction in the protocols and analysis of results. For example, a study by Petisco et al. (2019) (held after distinction of PAP and PAPE in 2017) concluded

that a moderate intensity like 80%-1RM back squat may induce greater improvements in explosive activities because of the PAP effect. Similarly, other authors evaluate the effects of PAP using protocols where performance in tasks was measured or whose evaluation time exceeds the time in which, according to the literature, the PAP phenomenon occurs.

In an attempt to explain why significant differences were not found in the values obtained in protocol B of this study (where there was no manipulation of the temperature and if it was intended to verify whether or not PAP was induced) it can be explained by several factor like, for example, (i) duration of MVC, it is been reported that contractions of 5-10 seconds produce the greatest PAP (Vandenboom, 2016), (ii) type of contractions, some evidences suggest that isometric stimuli may not be the most adequate to induce PAP (Seitz, 2016), still PAP effects on isometric strength have been considerably less studied, (iii) skin resistance and locations of the nerve relative to the stimulus (Palmieri et al., 2004) and, (iv) psychological state of subjects on the day of data collection (Zehr 2002).

Even if there is a gap in the current literature on this topic, Zimmerman et al., (2019) concluded that (i) the occurrence of PAP does not necessarily mean that the voluntary performance will be improved, (ii) improvement in voluntary performance is sometimes observed when the PAP level reaches extremely high values, (iii) and other mechanisms may be more relevant than for PAP in the manifestation of acute increases in performance following a conditioning contraction.

5.1.2. Relationship between temperature and the occurrence of the PAP phenomenon

When a study intends to analyse the effects of temperature on a physiological mechanism, thermoregulation processes (like shivering or sweating) must be taken into account because they are necessary to ensure optimal physiological function and survival. In a warm environment the skin temperature approaches the ambient temperature and the difference in temperature between the core and shell decreases as skins blood flow is increased (Tansey and Johnson, 2015). In the same study, it is explained that

during cold stress skin blood flow and temperature is reduced, leading to a conservation of heat to the core.

There are not many studies that relate the use of hot and cold packs to change the temperature in the soleus muscle and, much less, studies that relate temperature manipulation to the PAP phenomenon. Diaz et al. (2018) explained that the temperature of soleus muscle before applying the cold packs for 30 minutes, decrease in 43% and the largest decrease was in the first 5 minutes and the same happens when applied the hot packs. The 5 minutes of local cold and hot chosen were intended to lead to differences in the way in which the PAP mechanisms are activated by changing the temperature.

The results obtained after analyzing the values collected in the different temperature manipulation protocols in this study, a statistically significant difference in protocol A was found, showing an alteration in MVC values after cooling application which, through the observation of the graph 3, can be expressed by the decrease in performance. In the remaining variables, there were no significant differences after application of local cold or heat in the mechanisms of PAP activation.

Considering the limitations of this study and the fact that the results obtained in MVC, by themselves, do not justify changing the mechanisms associated with PAP, this value is the only one that meets the expectations since the evidence reported that isometric force was reduced with local cooling because of slowed nerve conduction velocity (for example) and with local heat the muscle fatigue parameters decreased related to muscular force production.

The lack of significant differences in the H-reflex variable under study can be explained by the reduced sample (N=5), the time that the individuals were exposed to the cold (=5 minutes with the cooling packs) and the fact that only the leg was cooled and not the entire lower limb/body leading to less contact area to stimulate thermoreceptors. Even though, as early as 2001, Rutkove stated that the cooling slows down the depolarization time while warming has the opposite effect and that’s why there is a facilitation of H-reflex output with cooling, Dewhurst et al. (2005) reported a 19% decrease in H-reflex after heat application. Long and Hopkins (2009) by applying 30 minutes of local heat did not find an

influence in H-reflex values or voluntary plantar-flexion peak to peak torque of the soleus muscle. These data show the existence of contrasting studies on this topic and the need for a more detailed exploration.

Despite the existence of a variable with significant differences, the results obtained are in agreement with Halder & Gao (2014) who report that the soleus muscle is known to be rich in blood volume, which may resist superficial cooling effects, and those of the study by Racinais (2013) which shows that warming the skin did not affect torque production or voluntary activation.

In the comparison between the variables under study in the second moment (post protocol), the existence of significant differences would prove the advantages of manipulating the temperature in the mechanisms of PAP activation. As there were no differences in any of the variables, in any of the protocols, considering the limitations of this study, the subjects did not benefit from the use of hot and/or cold.

It is argued that there is great inter-subject variability in the response to PAP and, that the values of MVC and H-reflex vary inter- and intra subject . For this reason, this study sought to ensure that the athletes were in a similar circumstances as possible, were from high explosive events, with at least 5 years of experience in strength training, and all attended the sessions before training in their respective clubs. Even though no significance differences were found between the subjects and, as can be seen in table 4, the subjects were in similar situations at the beginning of each data collection (pre-protocol).

From the thermophysiologic perspective, it is important that humans and other mammals have two types of skin: nonhairy and hairy and, temperature variation, as well as thermoregulation mechanisms are affected by the amount of hair around the segment under study since the two types, generally, have different temperatures (Romanovsky, 2014). Body fat has a strong influence on skin temperature, individual with high body fat composition hold heat in the body and the cooling process is slower (Webb, 1992), these are some examples of how individual characteristics change the way protocols act on subjects.

Following on from the above, and the fact that PAP is associated with high inter-individual variability, the lack of results under the effect of PAP on the

performance of athletes in explosive events of athletics and how temperature can influence the mechanisms associated with that phenomenon can also be explained by the various factors that lead to possible differences between subjects.

5.2. Study limitations

Critically analyzing the entire process inherent to this investigation, the limitations underlying a work of this nature are recognized, such as the number of subjects evaluated (N=5), which leads to a careful interpretation of the results and prudence in generalizations. This reduced number of subjects was due to different reasons, (i) being high-performance athletes, they could not perform activities that could impair performance, especially in the competitive season; (ii) the need to travel to the facilities where the samples were collected (LABIOMEP) and as well as to coordinate the schedules of these same facilities with those of the subjects; (iii) in addition, what led to a reduced number of subjects was that there were more non-responders (subjects whose values associated with electromyography were low and not detectable by the software) than expected. In future investigations, a strategy is suggested to allow the selection of larger and more representative samples, such as, for example, collecting data at the beginning or at the end of the sports season.

Other limitations were related to the scarcity of bibliography to support the discussion of the results obtained, given that there are few similar studies that allow comparison, and the use of skin temperature instead muscular temperature due to the lack of funding for the instruments needed to collet this possible variable.

Considering the mentioned limitations, this study leaves open the reflection on the importance of future investigations.

We now proceed to discuss the results obtained.

6. CONCLUSIONS

To contribute to the development of scientific knowledge, the main objective of this study was to verify if there was a relationship between the decrease or increase in temperature and the PAP phenomenon.

The results obtained cannot respond to the research question, since it is not possible to say that there is a change in the mechanisms of PAP activation under different temperature circumstances nor how the temperature manipulation protocols under the PAP effect can contribute to changes in sports performance.

Recognizing the inherent limitations of this study, after analyzing the data obtained in the different protocols, comparing the performance of the subjects in the MVC, variations of the H-reflex and the mean of the M-waves, no statistically significant results were found. Being that, it is important to be able to prevent the misconceptions between PAP and PAPE mechanisms and effects, so that there is uniformity in the way researchers, coaches, strength and conditioning coaches, among others, face these phenomena as a mean of increasing sports performance. Also, further studies are needed to understand whether the activation of PAP mechanisms is still advantageous in explosive modalities or if it has already become an "outdated phenomenon".

With regard to the findings on the effect of temperature on PAP, further studies are still needed to determine if cold and local heat applications genuinely modify the mechanisms of PAP activation with the exploration of different temperature manipulation methods and different exposure times to these instruments.

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8. APPENDIX

In this chapter, the informed consent term given to the subjects at the beginning of data collection and the sociodemographic data collection instrument used (questionnaire) are attached.

8.1. Informed Consent Term (portuguese version delivered to athletes)

TERMO DE CONSENTIMENTO INFORMADO

Título do Estudo: Influência da Temperatura nos Mecanismos que Induzem Potenciação Pós-Ativação: Um estudo Randomizado.

Identificação do Investigador

O meu nome é Joana Pinho Ferreira, sou mestranda em Treino Desportivo-Treino de Alto Rendimento, na Faculdade de Desporto da Universidade do Porto. O orientador da investigação é o Professor Doutor Filipe Almeida Viana da Conceição, da Faculdade de Desporto da Universidade do Porto.

Informações aos Participantes sobre o Estudo

No âmbito do mestrado, estou a realizar uma investigação cujo **objetivo** é explorar a relação entre temperatura e ocorrência do fenómeno PPA (potenciação pós-ativação*)*, bem como, identificar a relevância deste fenómeno para o aumento da *performance* em praticantes de disciplinas explosivas do atletismo.

Esta investigação tem como finalidade compreender de que forma a manipulação da temperatura sob os mecanismos associados ao fenómeno PPA, pode contribuir para o aumento da *performance* atlética e, consequentemente, o aumento dos resultados obtidos em competição.

A colheita de dados será realizada em contexto de laboratório, nas instalações do Laboratório de Biomecânica do Porto (LABIOMEP).

Caso dê o seu consentimento para a sua participação, serão respeitadas as seguintes condições:

1. A sua participação é voluntária e a recusa não implicará quaisquer prejuízos.
2. As informações que fornecer ao investigador serão utilizadas exclusivamente para fins científicos.
3. A sua identidade será sempre preservada e as respostas/dados recolhidos serão mantidos anónimos e confidenciais.

4. A desistência do compromisso de participação é livre em qualquer momento.
5. A qualquer momento, pode solicitar ao investigador informação adicional sobre a participação no estudo.
6. Caso queira contactar o investigador, pode utilizar o telefone 910411296 ou o e-mail joanapinhoferreira2@gmail.com.
Na posse da informação e nas condições acima expressas, declaro que aceito participar na pesquisa através da recolha de dados em laboratório, assumindo não ter sofrido qualquer pressão para tal.

Data __/__/____

Assinatura _____

Assinatura do Investigador _____
(Joana Pinho Ferreira)

8.2. Sociodemographic questionnaire (portuguese version delivered to athletes)

Questionário nº____

"INFLUÊNCIA DA TEMPERATURA NOS MECANISMOS QUE INDUZEM POTENCIAÇÃO PÓS-ATIVAÇÃO: UM ESTUDO RANDOMIZADO"

Este questionário enquadra-se numa investigação, proposta no âmbito do Mestrado em Treino Desportivo-Treino de Alto Rendimento a decorrer na Faculdade de Desporto da Universidade do Porto.

Para concretizar esta investigação necessitamos da sua preciosa colaboração. O questionário é **anónimo**, não devendo por isso colocar a sua identificação em nenhuma das folhas nem assinar o questionário. Não existem respostas certas ou erradas. Por isso lhe solicitamos que responda de forma sincera a todas as questões.

Agradeço, desde já, a sua colaboração e disponibilidade.

Com os melhores cumprimentos,

Joana Ferreira

CARACTERIZAÇÃO SOCIODEMOGRÁFICA

1. Idade _____ anos
2. Altura _____ cm
3. Peso _____ kg

CARACTERIZAÇÃO DESPORTIVA

1. Escalão
   _____
2. Anos de Prática
   _____
3. Modalidade e Disciplina Praticada
   _____
4. Anos de Experiência em Treino de Força
   _____

CARACTERIZAÇÃO CLÍNICA DA SITUAÇÃO DO ATLETA

1. Teve alguma lesão no desporto?

__ SIM __ NÃO

a. Se sim, indicar qual/quais e há quanto tempo.

_____
_____
_____

2. Toma regularmente algum medicamento ou outras substâncias?
__ SIM __ NÃO
   a. Se sim, quais?

_____
_____
_____

3. Tem alguma doença que considere relevante informar?
__ SIM __ NÃO
   a. Se sim, quais?

_____
_____
_____

4. Recentemente, foi sujeito a processo de eletroestimulação?
__ SIM __ NÃO
   a. Se sim, há quanto tempo?

_____

Muito Obrigado pela colaboração!