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Acute effects of a High-Intensity Functional Training (HIFT) session on muscle damage and recovery capacity of individuals with different fitness status

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Dissertação apresentada ao Programa de Pós-Graduação em Educação Física e Esporte da Escola de Educação Física e Esporte de Ribeirão Preto da Universidade de São Paulo para obtenção do título de Mestra em Ciências.
Área de concentração: Aspectos biodinâmicos da atividade física e do esporte.

Orientador: Enrico Fuini Puggina.

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DEDICATÓRIA

Consagro esta dissertação aos meus pais Ana Lúcia e Heraldo que me proporcionaram a oportunidade e o privilégio de dedicar minha infância e adolescência aos estudos, fazendo com que eu pudesse ingressar em uma Universidade, conquistar o título de bacharela em Educação Física e Esporte e, agora na pós-graduação, pudesse desenvolver o presente trabalho. Minha formação profissional e acadêmica é consequência da formação pessoal que adquiri ao longo da vida, tendo meus pais como exemplo de caráter e dedicação.

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“Permita que eu fale, não as minhas cicatrizes. Se isso é sobre vivência, me resumir à sobrevivência é roubar o pouco de bom que vivi.” (Emicida)

RESUMO

SILVA, A. E. S. **Efeitos agudos de uma sessão de Treinamento Funcional de Alta Intensidade (TFAI) no dano muscular e na capacidade de recuperação de indivíduos com diferentes estados de condicionamento físico.** 2023. Dissertação (Mestrado em Educação Física e Esporte) - Escola de Educação Física e Esporte de Ribeirão Preto, Universidade de São Paulo, Ribeirão Preto, 2023.

BACKGROUND: O Treinamento Funcional de Alta Intensidade (TFAI) é amplamente utilizado atualmente devido à baixa demanda de tempo e eficiência para melhorar o desempenho e a saúde. A dinâmica de recuperação do dano muscular e do condicionamento físico após um TFAI em indivíduos com diferentes estados de condicionamento físico fornecem informação prática para treinadores e praticantes. Portanto, o objetivo deste estudo foi verificar as respostas de dano muscular e recuperação de desempenho após uma sessão aguda de TFAI em homens jovens saudáveis com diferentes estados de condicionamento físico. MÉTODOS: Dezesseis participantes treinados recreacionalmente (idade: 23,4 ± 2,4 anos; índice de massa corporal: 24,6 ± 2,4 kg·m-2; uma repetição máxima [1RM] no agachamento: 120,1 ± 19,9 kg) foram divididos em dois grupos de acordo com sua força máxima (mais treinados [HT] e grupo menos treinado [LT]) e realizaram uma única sessão de TFAI. Dano muscular (creatina quinase [CK] e lactato desidrogenase [LDH]) e testes de aptidão física (força, potência e consumo de oxigênio) foram analisados antes, imediatamente após, 24h e 48h após a sessão de TFAI. A carga interna de treinamento para ambos os grupos foi equalizada por meio da Percepção Subjetiva de Esforço (PSE) e o percentual de 1RM. RESULTADOS: Marcadores bioquímicos mostraram que ambos os grupos sofreram dano muscular induzido pelo exercício, ao passo que o desempenho (altura de salto no *Countermovement Jump* (CMJ), Potência Relativa no CMJ para ambos os grupos e VO_2máx para o grupo LT) foram afetados imediatamente após a sessão de TFAI. Houve uma tendência de recuperação mais rápida do dano muscular no grupo HT. CONCLUSÕES: O grupo HT apresentou maior recuperação do dano muscular em comparação com o grupo LT. Pode-se esperar um tempo de recuperação mais longo para a recuperação muscular completa no grupo LT.

Palavras-chave: Fadiga. Aptidão física. Recuperação. Treinamento intermitente.

ABSTRACT

SILVA, A. E. S. **Acute effects of a High-Intensity Functional Training (HIFT) session on muscle damage and recovery capacity of individuals with different fitness status.** 2023. Dissertação (Mestrado em Educação Física e Esporte) - Escola de Educação Física e Esporte de Ribeirão Preto, Universidade de São Paulo, Ribeirão Preto, 2023.

BACKGROUND: High-Intensity Functional Training (HIFT) is nowadays widely used due to low time demand and efficiency to improve performance and health. The dynamics of recovery of muscle damage and physical fitness after a HIFT in individuals with different fitness status provide practical information for coaches and practitioners. Therefore, the aim of this study was to verify the muscle damage and performance recovery responses after an acute HIFT session in healthy young men with different fitness status. METHODS: Sixteen recreationally trained participants (age: 23.4 ± 2.4y; body mass index: 24.6 ± 2.4kg·m-2; one maximum repetition [1RM] back squat: 120.1 ± 19.9kg) were divided in two groups according to their maximum strength (higher-trained [HT] and lower-trained group [LT]) and performed a single HIFT session. Muscle damage (creatine kinase [CK] and lactate dehydrogenase [LDH]) and physical fitness tests (strength, power, and oxygen consumption) were analyzed before, immediately after, 24h and 48h after the HIFT session. The internal training load for both groups was equalized using the Rating of Perceived Exertion method (RPE) and the percentage 1RM. RESULTS: Biochemical markers showed that both groups suffered exercise-induced muscle damage, whereas performance (jump height in Countermovement Jump (CMJ), Relative Power in CMJ for both groups and VO_2max for LT group) were affected immediately after exercise HIFT session. There was a tendency for faster muscle damage recovery in HT group. CONCLUSIONS: HT group showed higher muscle damage recovery compared with LT group. Longer recovery time might be expected to complete muscle recovery in LT group.

Key words: Fatigue. Physical fitness. Recovery. Intermittent training.

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SUMMARY

1 INTRODUCTION

High-Intensity Interval Training (HIIT) is a time-efficient training method widely used due to the capacity of improving health and performance in a brief demand of time (Feito; Brown; Olmos, 2019), once it is possible to accumulate more stimuli in a shorter period of time given its characteristics. Recently, research on HIFT reported high energy expenditure, improvements in body composition and absolute strength (Bahremand; Hakak Dokht; Moazzami; 2020 and Browne *et al.*, 2020). It involves repeated short (lower than 45 seconds) to long (two to four minutes) bouts of high intensity exercises interspersed with recovery periods (Buchheit; Lauresen, 2013). The periods of efforts require near maximal, maximal or "*all-out*" efforts. The high intensity could be expressed analyzing variables such as heart rate (HR), oxygen consumption (VO_2max), rate of perceived exertion (RPE) or strength since the values reach ≥ 80% of maximal capacity (*e.g.*, as a strategy to determine high intensity) (Buchheit; Lauresen, 2013). A HIIT program consists of the manipulation of the following variables (relying on the individual fitness): number of sessions per week, number of bouts per session, intensity of bouts, duration of each bout, recovery between each bout (related to duration) and type of recovery between each bout (passive rest or low-intensity exercise) (Buchheit; Lauresen, 2013).

In general, HIIT involves cyclic exercises, such as running, cycling or rowing. Recently, High Intensity Functional Training (HIFT) or High Intensity Cardioresistance Training (HICRT) are described as an alternative of HIIT for "functional" exercises, mostly performed with bodyweight or even using external load to increase intensity (Buchheit; Lauresen, 2013). According to Greenlee *et al.* (2017) and Menz *et al.* (2019) there are conceptual commonalities with HIIT (*i.e.*, high intensity nature of training), however they differ from traditional HIIT because they emphasize multi-joint movements that enhance not only the aerobic and cardiovascular capacity but also muscle power and strength (Greenlee *et al.*, 2017 and Menz *et al.*, 2019). It leads to the conclusion that this peculiarity allows different physiological responses and anaerobic adaptations, since the functional training involves endurance and resistance-based patterns of movement (Feito *et al.*, 2018). This concept matches the popular CrossFit® training strategies, which provide multiple training benefits within the same session (Schlegel, 2020).

High intensity training leads to exercise-induced muscle damage (MD) and post-exercise fatigue (Mate-Munoz *et al.*, 2017 and Heavens *et al.*, 2014). The MD comprises in structural disruptions as a result of mechanical strain and metabolites accumulation that culminates in efflux of muscle enzymes – such as Creatine Kinase (CK) and Lactate Dehydrogenase (LDH), and in performance loss (decrease of strength and power) (Bishop; Jones; Woods, 2008 and Tibana *et al.*, 2016). In recent decades, many studies have shown a strong relationship between high intensity training and MD (Takahashi *et al.*, 1995). Gomes *et al.* (2020) investigated MD following a single 'Cindy' workout session (classified as HIFT) in adult practitioners. The workout elicited significant acute perturbations in the analyzed muscle cells by increased CK activity after the exercise bout (174.9 to 226.7 U.L-1), that remained elevated after 24 hours after training session completion. Ertel, Hallam and Hillman (2020) analyzed the effects of training status and exercise intensity on exercise-induced MD. They found that high intensity exercise results in greater MD in both trained and untrained individuals. Moreover, higher-trained people seem to have less pain and recover faster, although their CK levels are higher.

Recently, Tibana *et al.* (2022) revealed that eight recreational male participants increased their CK levels one hour after a HIFT workout on participants that performed all-out intensity while those who performed moderate intensity HIFT workout (rating perceived exertion 6 [RPE]) did not increased their CK levels as much as the all-out intensity group. The training intensity and the low rest intervals are the key factors of the muscle damage (Tibana *et al.*, 2019). Tibana *et al.* (2019) reported that nine HIFT athletes tended to increase their CK levels after three days of a HIFT with the CK values decreasing only 72 hours after HIFT workout. According to the authors, this phenomenon was able to affect the performance of the participants. Although the HIFT compromises metabolic, gymnastic and weightlifting exercises, the weightlifting exercises performed at high intensity (multiple repetitions and sets) demonstrate to impair performance more than the other variables (i.e., metabolic and gymnastic exercises) (Mate-Munoz *et al.*, 2018). The power clean and the snatch movements are common in HIFT-workouts. Usually, the intensity performed is submaximal, and due to this fact, the movement speed and repetitions are higher. Mate-Munoz *et al.* (2017) described that maximum repetitions of power clean at 40% 1RM increased the lactate levels more than 10-fold. Thus, it is expected that HIFT-workouts comprising weightlifting exercises lead to higher training loads than metabolic movements, also resulting in longer recovery periods to restore performance (Mate-Munoz *et al.*, 2017).

According to the general theory of training, the decrease in functional status after training is a prerequisite for adjustments to the stimulus to occur (Chiu; Barnes, 2003). The Fitness-Fatigue model associates stress arising from training with physiological responses. Among all the factors that can affect performance and recovery after a training session, the fitness status and fatigue stand out. The first one is a positive physiological response, whereas the second one is negative. The effects of fatigue are related to neural and metabolic systems. Decreases in nervous system function and in storage and availability of energy substrates are the main reasons why performance is impaired. Recovery after training stimulus is necessary to reverse the negative impacts caused by the fatigue process, allowing performance improvement (Chiu; Barnes, 2003). The recovery period is individual-dependent, based on fitness status. Well-trained individuals are protected from MD, since mechanisms acquired by skeletal muscles prevent and provide less exercise-induced injuries (Repeated Bout Effect - RBE- phenomenon) (Mchugh *et al.*, 1999). Another explanation is related to an augment in efficacy of the contractile components of the muscle in well-trained individuals, leading to less stress being placed and reduced MD (Johnston; Gabbett; Jenkins, 2015), allowing the recovery from training within 48h (Sjökvist *et al.*, 2011).

Post-exercise recovery is a fundamental component for continuous performance enhancement. The recovery from training stimuli and exercise-induced adaptations take place between sessions. Recovery after training is chiefly a matter of reversing the MD and fatigue to restore homeostasis in the muscle cell (Bishop; Jones; Woods, 2008). The negative impact of neglecting adequate recovery and performance in fatigue status is directly related to overtraining (Kenttä; Hassmén, 1998) and risk of injury. The exercise stimuli leads to fatigue that in turn decreases performance (*e.g.*, muscle strength, power) (Maté-Muñoz *et al.*, 2018). This weariness after training negatively impacts the ability to perform a new training session, compromising physical fitness improvement. The recovery after the HIFT workout is necessary to reverse the negative impacts caused by the fatigue process allowing performance improvement (Chiu; Barnes, 2003). Previous studies on MD time recovery reported that 24h after HIFT workout was not sufficient to reestablish the muscle structure and function. Interestingly, none of those studies investigated the MD time recovery between individuals from different training levels aiming to understand the dynamics of recovery of performance and MD.

Despising the fact that HIFT is capable to increase lactate more than 10-fold, as well as CK activity, it is also recognized in the literature various key points related to the physiological effectiveness of training, such as the improving physical capacity, including aerobic and anaerobic capacity, anaerobic power, cardiovascular fitness, body fat reduction, and the resistance to sustain high lactate levels for high-volume training (Gianzina; Kassotaki; 2019). Recently, Meier, Sietmann and Schmidt (2022) reported that participants of different training levels who trained HIFT achieved similar HR behavior, and this is possible due to the HIFT design, combining aerobic and anaerobic exercise intensities. Despite this fact, the more experienced HIFT participants are able to withstand higher loads (i.e., heavier loads [kg]). In this sense, studies in which the effects of HIFT on MD and performance among subjects with different training levels are scarce.

Thus, we aimed to investigate MD exercise-induced and recovery through biochemical markers and performance indicators according to fitness status to a better understanding about the time-course of recovery in higher and lower trained subjects. As mentioned before, several studies were designed to investigate the traditional HIIT, while the high intensity resistance-based exercises are still misunderstood regarding MD and recovery. Moreover, the investigations should embrace and consider the influence of individual training status whereas higher and lower-trained individuals demonstrate different outcomes from training. Based on the previous literature (Barbieri *et al*., 2019 and Messonnier *et al*., 2013), we hypothesized that higher trained participants should experience less MD and recover quickly when compared with lower trained participants exposed in the same training protocol.

2 LITERATURE REVIEW

“How is fatigue established and how does recovery behave after HIIT or HIFT training according to the fitness status?” In literature there are many findings about physical exercise and its consequences. Physical exercise affects the balance of the internal environment, since during the execution of movements, muscle contraction generates disturbances in the homeostasis of the human body. According to Ament and Verkerke (2009) the production of force, power and heat during exercise manifests a form of mechanical energy. This mechanical energy will utilize the body's energy stores, generating metabolites and heat that affects the steady state. Such disturbances initiate sensations of tiredness and fatigue and the physiological role of this sensation is to protect the individual from the deleterious effects of exercise. Muscle cells have their biochemical balance affected by exercise (inorganic phosphate, hydrogen protons, lactate, among others are accumulated), which affects the mechanical machinery. Additionally, influences the process of neuronal signals transmission by the organelles involved. Fatigue, therefore, is the definition of the physiological effects caused by exercise at the physical and biochemical levels (Ament; Verkerke, 2009).

During the process of muscle contraction, many metabolites are generated due to energy demand by cells (ATP). Such metabolites influence the homeostasis of the system and establish what is called peripheral fatigue. When this happens, it impairs the coupling of actin and myosin and consequently affects muscle contraction. The accumulation of inorganic phosphate in the sarcoplasm decreases the force of contraction due to the inhibition of the cross-bridge interactions. In addition, it also affects calcium reuptake by the sarcoplasmic reticulum. The decline of peripheral glucose stores (glycogen) and blood glucose levels interfere with the functions of the central nervous system, thus, the peripheral fatigue mechanism can be understood as a protection strategy for the organism. Furthermore, the flow of energy from the fibers can lead to a blockage in excitation-contraction signaling. During exercise, there is also an increase in the respiratory quotient in an attempt to decrease the hydrogen concentration. There is an accumulation of ammonia due to heat production, leading to increased sweating and water loss, which can culminate in dehydration (Ament; Verkerke, 2009).

Also according to Ament and Verkerke (2009), central fatigue can be understood by the synaptic effects of serotonergic neurons, causing the feeling of tiredness and fatigue (which can be explained by the increase in serotonin in the brain). The decrease in aminoacid concentration and the increase in cytokines also induce the feeling of fatigue during exercise. Interleukin-6 (IL-6) released during exercise can be 50 times higher than baseline during resting conditions.

In addition, the muscle damage caused to the fibers introduces a post-exercise inflammatory process. All these system disturbances generate impairments that after training will need to be repaired.

In a study by Alba-Jiménez, Moreno-Doutres and Peña (2022) neuromuscular fatigue is defined by the exercise-induced reduction in the maximum voluntary force that the muscle can generate. The change in neuromuscular function (NMF) occurs due to muscle contraction and can be understood by peripheral and central fatigue, being detected for up to 48 hours after exercise. Peripheral fatigue develops earlier at the neuromuscular junction and therefore performance is influenced. Central fatigue appears after voluntary neural muscle activation and tends to occur later. Work accumulation and incomplete strength restoration can influence performance, and therefore, fatigue management is essential to control athletes' adaptations and reduce susceptibility to injury. Some strategies to monitor and describe fatigue are questionnaires, biochemical markers, sprint tests and vertical jump tests. From a subjective point of view, Rating of Perceived Effort (RPE) has been widely used because of its simplicity and high validity (Borg, 1998; Foster, 2001). Regarding objective measures, the Counter Movement Jump (CMJ) is a tool of high validity and reliability.

Fatigue can be understood as a reduction in the ability to obtain the desired performance, which limits physical and cognitive function. Short term fatigue may have a metabolic origin, while prolonged fatigue originates at a neuromuscular level. If fatigue is sustained and recovery strategies are not correct, athletes may be at risk of performance decline and, even worse, injury. Although the control of NMF is essential, the period needed to recover it is not well established. It has been reported in the literature that 24 to 48 hours of recovery are necessary to return to performance capacity in some jump tests (Doeven, 2018). In other findings this period can reach up to 72 hours. It is important to note that recovery has an individual component that must be taken into account. Despite this, aerobic metabolism (*e.g.* fitness status) plays an essential role, especially after high-intensity training, to restore homeostasis and minimize the drop in neuromuscular performance (Bishop; Jones; Woods, 2008).

In the last mentioned study from Bishop, Jones and Woods (2008), as in the present work, the biochemical markers of acute response were fundamental to determine the recovery time after exercise. Creatine kinase (CK) is a marker that is characterized by indicating muscle damage, as it stays inside the muscle cell, but usually overflows into the bloodstream after intense exercise. Vertical jump tests are a practical measure of neuromuscular ability and have been used to investigate the recommended recovery time after strenuous activity. These tests are more sensitive for determining fatigue as they reflect the stretch-shortening capacity of the lower limb muscles and the possibility of assessing muscle fatigue considering performance. The Squat Jump (SJ), Drop Jump (DJ) and CMJ are test options used, but CMJ is the most popular to assess fatigue due to its reliability and validity. It is considered the gold standard and is currently the most accurate test to detect neuromuscular fatigue.

To better understand fatigue, the study by Halson (2014) aimed to monitor the training load, since fatigue is influenced by the type of stimulus, type of contraction, duration, frequency and intensity of the exercise. Physiological assumptions, the athlete's training status, and environmental conditions also influence fatigue. Due to this multifactorial nature and the inherent complexity it is a challenge to monitor and measure fatigue in athletes. Monitoring an athlete's training load is seen as a determining factor to classify whether the individual is adapting to the training program (ensuring program efficiency and performance maintenance), minimizing the risk of overreaching, overtraining and injuries. Despite the importance of training load monitoring, a single definitive tool that is both accurate and reliable is not self-evident. Therefore, more than one tool is generally used, being again the biochemical markers and performance indicators (mainly vertical jump tests as CMJ).

According to Byrne, Twist and Eston (2004) exercise-induced muscle damage is a common phenomenon that results from exercising with high intensity and/or prolonged duration. Muscle damage is the state that can be identified by one or more indirect direct indicators. Documented symptoms include disfigurement of muscle cell structure, sarcolemma and extracellular components, compromising function and impairing performance. One of the most effective ways to assess the magnitude of muscle damage is through biochemical markers and functional consequences, such as reduced strength and power. Regarding the enzymatic markers of peripheral fatigue during exercise, Finsterer (2012) determined that exercise-induced fatigue and the indirect measurement of muscle damage through biochemical analysis depend on age, gender, training status, exercise mode and duration. Peripheral fatigue and the cessation of activity due to the impossibility of performing muscle contraction is a protection mechanism against the deleterious effects of exercise, but before it is established, there is damage to muscle cells. Especially in high-intensity exercises, which recruit type II fibers, there is an exacerbated activity of the CK enzyme, that efflux to the bloodstream. In addition, there is also the production of IL-6. Therefore, such biochemical markers are important to analyze the process of muscle damage and fatigue, especially in high-intensity exercises. Furthermore, it is important to consider that biochemical markers may return to baseline values, but fatigue may still be present. Hence, a way to assess fatigue assertively is comparing performance (the maximum capacity of a muscle) with such biomarkers, as proposed in this present work.

An original study by Wiewelhove *et al*. (2016) confirmed the onset of fatigue after a HIIT program in tennis players. The researchers investigated the effect of active and passive recovery on markers of fatigue. Eight tennis players (age 15.1 ± 1.4 years) internationally ranked between 59 and 907 (International Tennis Federation) participated in the study. After the HIIT session (3 sets of 8 bouts, with 20 seconds of passive recovery between bouts and 6 minutes of passive recovery between each set; each bout lasts 15 seconds and consists of 20 meters of shuttle runs at 90% maximum velocity reached in the agility test). Players completed 15 minutes of jogging or a passive recovery (this was a crossover study with a 4 month washout period). CMJ tests, CK concentration, delayed onset muscle soreness (DOMS) and perceived fatigue and stress were measured before and after the HIIT protocol. The results showed that high-intensity interval training induces a reduction in CMJ performance and an increase in CK activity. In addition to the increase in the perception of stress when comparing the post-intervention with the pre-intervention results. These findings allow to affirm the presence of muscle damage and the subsequent onset of fatigue due to the training protocol, but the active or passive recovery intervention did not culminate in significant differences.

In line with the last study mentioned, an original study by Verschueren *et al*. (2021) analyzed the interaction of acute fatigue with 3 traditional performance tests in 20 recreational athletes (age 24 ± 3 years). To induce the fatigue state a 30-second all-out effort was performed (adapted Wingate test). During the training protocol, there was a significant increase in heart rate, systolic pressure, lactate concentration and perceived exertion. Performance on single leg hop tests for distance (SLH), Counter- Movement Jump (CMJ) and Y-balance test (YBT) was impaired after the protocol, stating that high-intensity training can introduce fatigue. Howatson and Milak (2009) also tried to elucidate the exercise-induced muscle damage and fatigue after a sprint match. For such, 20 male subjects (age 22,6 ± 2 years) performed 15 sprints of 30 meters with an interval of 60 seconds. Maximum isometric strength, CK activity and muscle soreness were analyzed before, 24, 48 and 72 hours after the training protocol. The results showed significant effects of training on muscle damage for all variables mentioned. CK activity and soreness were significantly greater above baseline by 72 hours after exercise, while isometric strength was lower only 48 hours after exercise. These data elucidate that a high-intensity interval training protocol generates muscle damage and impairs performance on subsequent days.

To elucidate the period of time necessary for complete recovery, Twist and Eston (2005) analyzed the effects of exercise-induced muscle damage on performance during maximal intensity intermittent exercise. As already well described, muscle damage impairs athletic performance, especially when it comes to maximum intensity exercise. The objective of the study was to analyze the effects of this muscle damage on the maximum performance of the intermittent sprint. For such, 10 male participants (age 22.4 ± 3.2 years) performed a sprint in the cycle ergometer of 10.6 seconds interspersed with 24 seconds of recovery against a load of 0.10 kp/kg and 10 x 10 m sprints from a standing start, each with 12 seconds of active (walking) recovery. The variables were measured before, 30 minutes, 24, 48 and 72 hours after a plyometric exercise protocol comprising of 10x10 maximal CMJ. The analyzed variables showed changes in perceived pain, CK activity, peak power, sprint time and rate of fatigue. Pain was greater than baseline at all intervals as well as CK activity at all post-test time points compared to baseline. Peak power was also lower and the fatigue rate was higher, but with a 40% reduction occurring after 48 hours. After 72 hours all variables analyzed returned to baseline. The results show that after the onset of muscle damage induced by plyometric exercise, the ability of the muscle to generate energy is reduced for at least 3 days.

To investigate routines for fatigue analysis in HIIT, Wiewelhove *et al*. (2015) assayed fatigue recovery in men and women from sports teams during high-intensity interval training. The aim was to investigate changes in fatigue and recovery markers in response to HIIT and the assessments routines. 22 athletes (age 23.0 ± 2.7 years) practiced a six-day microcycle with 11 sessions. Repeated-sprint ability (criterion measure of fatigue and recovery), CMJ height, 20 meters sprint performance, muscle contractile properties, serum activity of CK and perceived muscle soreness (DOMS) were measured pre and post the training program as well as after 72 hours of recovery. The results proved that all the variables mentioned before were impaired by HIIT training, which demonstrates significant changes in performance and also the presence of fatigue. In addition, all tests can be used as indicators of fatigue and recovery, but the combination of assessments allows for a more assertive conclusion.

Fitzpatrick *et al*. (2019) in order to analyze the reliability of fatigue monitoring measures of elite youth soccer players applied a well-being questionnaire, jump performance test and accelerometer variables during submaximal shuttle running in 17 male players (age 17.4 ± 6.5 years). The evaluation occurred on two occasions with an interval of seven days and the results suggested that the CMJ has good reliability. The subjective questionnaire showed low reliability. The findings provide information about the reliability of available tools to measure and monitor fatigue.

It’s well known that fatigue status can culminate in the overtraining syndrome. In another original study, authors Cadegiani, Kater and Gazola (2019) investigated the hormonal and metabolic consequences of HIIT training regimes and the overtraining syndrome. They recruited healthy CrossFit® practitioners diagnosed with overtraining and a sedentary control group. CrossFit® athletes presented increased cortisol, neutrophils, testosterone, basal metabolic rate (BMR) ratio and fat oxidation. Conversely, more than 90% of the adaptive changes in athletes were lost under overtraining syndrome.

Many researchers tried to elucidate strategies to speed up recovery. Vanderlei *et al*. (2017) analyzed post-exercise recovery through the investigation of metabolic, clinical and biological variables after different cold water immersion (CWI) temperatures and durations. 105 male subjects were recruited, divided into a control group and four recovery groups. The volunteers were submitted to an exhaustion protocol consisting of a jump program and the Wingate Test and right after were directed to a tank with water and ice. Blood samples were collected for later lactate and CK analysis pre, 24, 48, 72 and 96 hours after the training protocol. For CK the duration of 15 minutes at 14° was the best option considering the results of 72 hours after exercise and for lactate 5 minutes at the same temperature. The results for perception of recovery demonstrate 5 minutes at 14 °C (96h) was the best strategy. For pain, no one of the interventions were effective. Despite that, in conclusion, there are no significant differences between recovery and control groups. Thus, passive rest and adequate nutritional support is still the best recovery strategy.

As mentioned before, recovery period is individual-dependent and partially based on fitness status. The Repeated Bout Effect -RBE- phenomenon partially explains why well-trained individuals recover faster. They are protected from MD, since mechanisms acquired by skeletal muscles prevent and provide less exercise-induced injuries (Mchugh *et al*., 1999). Another explanation is related to an augment in efficacy of the contractile components of the muscle in well-trained individuals, leading to less stress being placed and reduced MD (Johnston; Gabbett; Jenkins, 2015). Despite that, it still remains unclear how fatigue is established and how recovery behaves after HIIT or HIFT training according to the fitness status.

3 OBJECTIVES

3.1 GENERAL OBJECTIVE

We aimed to investigate exercise-induced MD and recovery responses through biochemical markers (CK and LDH) and performance indicators (physical tests of strength, power, and indirect oxygen consumption) after an acute HIFT session in healthy young men according to their fitness status.

3.2 SPECIFIC OBJECTIVES

The following stand out:

- Measure the participants' maximum strength using the One Maximum Repetition (1RM) in the back squat exercise (before, immediately after, 24 hours and 48 hours after the session) and also to calculate and define the work overloads in the session and even to determine the relative strength (in both groups);

- Measure the maximum power of the volunteers through the vertical jump test (Squat Jump [SJ]; Counter Movement Jump [CMJ]; Drop Jump [DJ]) before, immediately after, 24 hours and 48 hours after the session;

- Evaluate the maximum cardiorespiratory fitness from the incremental test on a treadmill to estimate the maximum oxygen consumption (VO_2max) before, immediately after, 24 hours and 48 hours after the session;

- Investigate the behavior of the plasma activity of Creatine Kinase (CK) and Lactate Dehydrogenase (LDH) before, immediately after, 24 hours and 48 hours after the session.

4 MATERIAL AND METHODS

Participants: Sample size was obtained by calculating sample power through the number of observations, using the article by Johnston *et al*. (2015), considering variables in common with this study, such as CK and lower-limb power measured through Counter Movement Jump (CMJ), guaranteeing a statistical power of 0.95 and alpha of 5% (software G*Power – Dusseldorf, Germany). Sixteen participants (healthy physically active young men aged 18 to 30) were divided in 2 equal groups (8 each): higher-trained (HT) and lower-trained (LT) according to lower-limb Relative Strength (for squat exercise) and training history (physical activity practice at least for 1 year for the first group, with training frequency maintained, and practice time not determined for the second group, with non-systematized physical training). The recommendation was to avoid changes in dietetic parameters and no extra exercise during the tests. Individuals with pre-existing diseases or injuries were excluded from the protocol. They signed the free and informed consent approved by the Local Research Ethics Committee (Nº: 73304717.0.0000.5659). All the experiments are according to the current legislation (Helsinki declaration).

Experimental design: Participants attended to pre-established local 5 times. At the first contact - day 0 (D0), anthropometric measurements were taken and they performed the 1RM, vertical jump and VO_2max tests, as well as a partial HIFT workout to be familiar with the training bout. Forty-eight hours after de D0, the participants performed the pre-test (D1). Four different procedures were done: 1. Blood collection; 2. Vertical jumps - Squat Jump (SJ), Counter Movement Jump (CMJ) and Drop Jump (DJ); 3. 1RM; 4. Incremental treadmill test. Forty-eight hours after D1, on day 3 (D3), they performed the proposed HIFT workout and, immediately after it, all tests mentioned before were carried out in the same order described ("post-test"). All the procedures were repeated after 24h and on 48h after the HIFT session (Table I).

Table I. Experimental design

Day	*Procedures*
Monday (D0)	Data collection Questionnaires Movements familiarization
Tuesday	Off
Wednesday (D1)	Blood collection Performance tests
Thursday	Off
Friday (D3)	HIFT workout Blood collection Performance tests
Saturday (D4)	Blood collection Performance tests
Sunday (D5)	Blood collection Performance tests

Note: D0, day 0; D1, day 1; D3, day 3; D4, day 4; D5, day 5; HIFT, high intensity functional training.

Variables analyzed: The variables analyzed to determine muscle damage (MD) and recovery behavior were 1. Enzymatic activity of Creatine Kinase (CK) and Lactate Dehydrogenase (LDH) and 2. Physical performance (strength, power and estimative of maximal oxygen consumption). These data allowed the evaluation of acute effects of HIFT session. Besides that, sample division (HT and LT) enables the investigation of fitness influence on the recovery process after the intervention. Plasma creatine kinase and lactate dehydrogenase activity were determined using Bioliquid® (Pinhais, Brazil) commercial kits, according to the method proposed by the manufacturer.

Strength test: To measure maximal strength, the 1RM test was performed for the back-squat exercise. The back-squat exercise started with the barbell on the rack. The subjects positioned the barbell on the shoulder with extended wrists and flexed elbows. Subjects' feet were shoulder-width apart, with toes pointing forward and slightly outward. Subjects squatted at a knee angle of approximately 120° and then fully extended their hips and knees. The exercise was examined through visual inspection by an experienced instructor who had 10 years of coaching experience with weightlifters (Pierce, 1997). All the participants had experience with the protocol. Initially, to perform a warm-up, the participants do a stimulus of 5 to 10 repetitions at 40-60% of the predicted maximum load; 1 minute of passive interval; the second stimulus of 3 to 5 repetitions at 60-80% of the predicted maximum load; passive 2 minutes rest; the third stimulus of 2 to 3 repetitions with 90% of the predicted maximum load. Finally, after a passive rest (3 to 5 minutes), the participants had their first attempt to perform the 1RM. If the practitioner performed more than one complete repetition (eccentric and concentric phase) in the final test phase, the attempt was repeated after a 3 to 5 minutes interval. If the 1RM was not obtained in 3 attempts the participant repeated the protocol after an interval of 48 hours (ACMS, 2014).

Power test: To verify lower limb power the groups performed three types of vertical jumps - *Squat Jump* (SJ), *Counter Movement Jump* (CMJ) and *Drop Jump* (DJ) at *Ergo Jump Platform* (Cefise®, Nova Odessa - Brazil). SJ: standing on the mat, with feet parallel right under the shoulders and hands on waist, the participant performed a vertical jump starting from the half squat position. CMJ: following the same position before the participant started from the upright position to perform the simultaneous knee and hip flexion and extension, for the subsequent performance of vertical jump. DJ: falling from the top of a box (50 centimeters) to the mat, trying to get out of the mat as soon as the feet touched it and avoiding hip or knee flexion. There were 3 attempts for each jumping technique (SJ, CMJ and DJ), with intervals of 1 minute between attempts of the same technique and 5 minutes between different techniques (Smirniotou *et al.*, 2008). For statistical analysis, it was considered the best jump between the three attempts of each jumping technique.

Estimative of maximal oxygen consumption (VO_2max): Both groups performed the incremental test on a treadmill using the Ellestad protocol (Ellestad, 1975). The test started with 3 minutes of walking at 2.7 km/h at a 10% grade, followed by three stages 2 minutes each at 4.8; 6.4 and 8 km/h; 3 minutes of running at 8 km/h at 15% grade, followed by 2 minutes at 9.7; 11.3; 12.9; 14,5; 16.1; 17.7; 19.3; 20.9; 22.5 km/h or until fatigue. Thus, from time (t) in minutes obtained, the estimative of VO_2max was established by equation (Ellestad, 1975): VO_2max (ml·kg-1·min-^{1}) = 4,46 + (3,933 * t).

HIFT workout: The training load for both groups was equalized by RPE-8 on a scale from 0 to 10 (Category-Ratio Scale - CR10) (Borg, 1998; Foster, 2001), and through 1RM for squat exercise (70% 1RM). Participants performed a standardized warm up protocol walking on a treadmill at 6 km/h for 5 minutes followed by 3 sets of 2 mobility exercises lasting 6 minutes (1. adduction and abduction; 2. flexion and extension of hips and shoulders simultaneously) during 40 seconds with 20 seconds of passive recovery between exercises and series. After three minutes of rest, the HIFT session composed by 3 sets of Back Squat (70%

1RM); Shoulder Press (self-determined load following RPE-8); Burpee; Abdominal Sit-Up for 40 seconds (at self-selected close to maximal intensity considering RPE 8) with a passive recovery of 60 seconds between each exercise and series was performed. Table 2 summarizes such information of the HIFT protocol.

Table II. Warm up, mobility and HIFT workout structure

	EXERCISES	**DURATION (t)**	**INTENSITY**	**REST (t)**	**TOTAL (t)**
WARM UP	Treadmill walk	5’	6km/h	-	5’
MOBILITY (3 sets)	Adduction and abduction[1]	40’’	RPE - 4	20’’	-
	Flexion and extension[1]	40’’	RPE - 4	20’’	-
	TOTAL	4’	-	2’	6’
	PASSIVE REST				3’
HIFT SESSION (3 sets)	Back Squat	40’’	70% 1RM	60’’	-
	Shoulder Press	40’’	Self-selected load (RPE - 8)	60’’	-
	Burpee	40’’	RPE - 8	60’’	-
	Sit Up	40’’	RPE - 8	60’’	-
	TOTAL	8’	-	11’	19’
	WARM UP + MOBILITY + HIFT SESSION				33’

[1]hips and shoulders simultaneously.

Note: RPE, Rating perceived exertion; t, time. (‘), minute/s; (“), seconds; RM, maximum repetition.

Statistical analysis: Statistical analysis was performed using IBM SPSS Statistics software, version 22.0 for Windows. The normality of the data (Shapiro-Wilk) and the Mauchly’s sphericity test were verified, and violation was detected for the sphericity test. The repeated measures of ANOVA (group x time) were used after the univariate analysis for correction. Repeated measures of ANOVA were computed to determine possible differences between and within the values of CK and LDH activities, jump performance, maximum strength, and aerobic power as a function of the groups analyzed and evaluation of the times pre- and post-HIFT workout. Bonferroni post-hoc test was used. To characterize the size of the effects, the Partial eta-squared (ηp2) was also computed (Cohen, 1988). The significance level was pre-fixed at 5% ($p < 0.05$).

5 RESULTS

The comparison between groups showed differences when comparing the biochemical markers. For CK, in the post-test vs. 48h and for LDH in the pre-test vs. 48h; post-test vs. 48h; 24h vs. 48h. Our findings showed significant differences between groups when comparing performance for 1RM (kg) values at all time points and also for Relative Strength (u.a.). When analyzing groups separately we found the following: the Height (cm) and Relative Power (W·kg-1) from CMJ reduced right after HIFT bout for both groups in the pre-test vs. post-test. Only LT reduced the oxygen consumption right after HIFT bout (pre-test vs. post-test).

Participants: Characteristics of participants and groups are exposed on Table III.

Table III. Characteristics of participants and groups

	HT (n = 8)	*LT (n = 8)*	*p* (η_p^2)
Variables	**Mean ± SD**	**Mean ± SD**	**Between group differences**
Age (years)	24,62 ± 4,03	22,25 ± 2,96	0.201 (0.62)
Height (cm)	173.1 ± 4.61	177.50 ± 4.65	0.080 (0.94)
Body mass (kg)	74.88 ± 10.89	76.43 ± 6.17	0.733 (0.17)
Load on 1RM (kg)	137.25 ± 29.45	103.00 ± 10.41*	0.008 (1.55)
Relative Strength[1] (a.u.)	1.82 ± 0.23	1.34 ± 0.79*	0.001 (2.82)

Note: HT, higher trained; LT, low trained; cm, centimeters; kg, kilogram; RM, maximum repetition; a.u., arbitrary units; *significant differences from HT group squat exercise.

CK: CK activity was only significantly increased comparing pre-HIFT workout between groups (eta = 0.10; p = 0.043). Although this result, CK increased 52% comparing pre-HIFT workout for HT group vs. post-HIFT workout LT group (eta = 0.98; p = 0.347); 65% comparing 24h post-HIFT workout LT group vs. pre-HIFT workout HT group (eta = 0.75; p = 0.589); 86% comparing 48h post-HIFT workout LT group vs. pre-HIFT workout HT group (eta = 0.89; p = 0.769); 62% comparing post-HIFT workout HT group vs. post-HIFT workout LT group (eta = 0.37; p = 0.328); 32% comparing 24h post-HIFT workout LT group vs. post-HIFT workout HT group (eta = 0.44; p = 0.795): 48% comparing 48h post-HIFT workout LT group vs. post-HIFT workout HT group (eta = 0.98; p = 0.396); 62% comparing 24h post-HIFT workout HT group vs. 24h post-HIFT workout LT group (eta = 0.72; p = 0.961); 35% comparing 24h post-HIFT workout HT group vs. 48h post-HIFT workout LT (eta = 0.42; p = 0.948); and finally, 30% comparing 48h post-HIFT workout HT group vs. 48h post-HIFT workout LT group (eta = 0.66; p = 0.991). Figure 1a shows the time course of CK activity between and within groups.

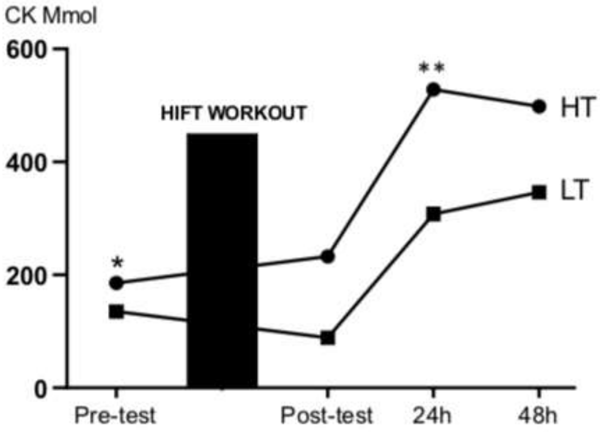

Figure 1a. Time course of CK activity pre- and post-HIFT workout.
Note: CK, creatine kinase; HIFT, high-intensity fitness training; HT, higher trained group; LT, lower trained group; *, difference between groups (pre-test); **, difference within group to post-test.

LDH: LDH did not changed between or within groups pre-, post-, 24h post- or 48h post-HIFT workout (n.s.). Although these results, LDH increased 5.5% comparing pre-HIFT workout HT group vs. pre-HIFT workout LT group (eta = 0.23; p = 0.847); 11% comparing pre-HIFT workout HT group vs. post-HIFT workout LT group (eta = 0.43; p = 0.792); 7% comparing 24h post-HIFT workout LT group vs. pre-HIFT workout HT group (eta = 0.23; p = 0.922); 41% comparing 48h post-HIFT workout LT group vs. pre-HIFT workout HT group (eta = 1.35; p = 0.526); 7% comparing post-HIFT workout HT group vs. post-HIFT workout LT group (eta = 0.30; p = 0.648); 11% comparing 24h post-HIFT workout LT group vs. post-HIFT workout HT group (eta = 0.37; p = 0.692): 47% comparing 48h post-HIFT workout LT group vs. post-HIFT workout HT group (eta = 1.53; p = 0.484); 8% comparing 24h post-HIFT workout HT group vs. 24h post-HIFT workout LT group (eta = 0.37; p = 0.376); 21% comparing 48h post-HIFT workout LT group vs. 24h post-HIFT workout HT (eta = 0.78; p = 0.921); and finally, 6.5% comparing 48h post-HIFT workout LT group vs. 48h post-HIFT workout HT group (eta = 0.22; p = 0.638). Figure 1b shows the time course of LDH activity between and within groups.

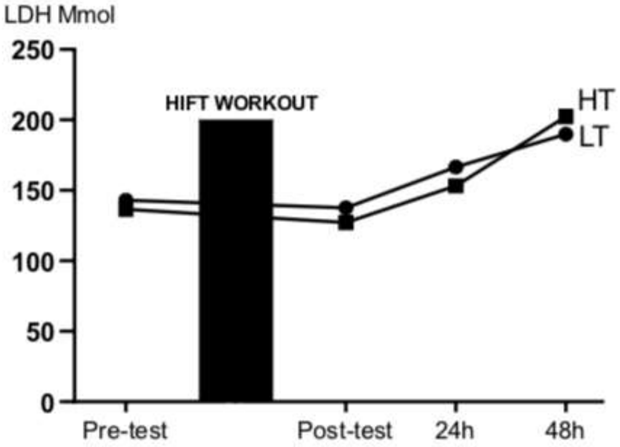

Figure 1b. Time course of LDH activity pre- and post-HIFT workout.
Note: LDH, lactate dehydrogenase; HIFT, high-intensity fitness training; HT, higher trained group; LT, lower trained group.

Fitness Performance: Our findings showed significant differences between groups for 1RM at all time points comparing HT vs. LT in pre-test (eta = 0.40; p = 0.008); post-test (eta = 0.37; p = 0.012); 24h (eta = 0.42; p = 0.006) and 48h (eta = 0.46; p = 0.013) and also for Relative Strength comparing HT vs. LT in pre-test (eta = 0.65; p < 0.001); post-test (eta = 0.60; p < 0.001); 24h (eta = 0.66; p < 0.001) and 48h (eta = 0.58; p = 0.001). There were no differences for moments in both groups when vertical jump and relative power in "W·kg-1" were measured on SJ, CMJ and DJ and for estimative of oxygen consumption (n.s.). However, significant differences were observed for the vertical jump in "cm" from CMJ reduced post-HIFT workout for both groups, demonstrating lower limbs fatigue, pre-HIFT workout vs. post-HIFT workout for HT group (eta = 1.11; p = 0.024) and LT group (eta = 0.80; p = 0.017) and from relative CMJ values comparing pre-HIFT workout vs. post-HIFT workout for HT group (eta = 1.11; p = 0.035) and LT group (eta = 0.76; p = 0.019). LT group reduced the VO_2max right post-HIFT workout, pre-HIFT workout vs. post-HIFT workout for LT group (eta = 1.21; p = 0.001). Data pertaining to the performance variables are exposed in table IV.

Table IV. Performance parameters at pre- and post-HIFT workout

	Pre-test		**Post-test**		**24h**		**48h**	
	HT	**LT**	**HT**	**LT**	**HT**	**LT**	**HT**	**LT**
Height SJ (cm)	36.4 (3.86)	34.65 (4.15)	33.5 (7.00)	32.1 (4.42)	34.35 (4.93)	34.6 (4.22)	33.45 (6.61)	32.9 (5.44)
Height CMJ (cm)	39.5 (5.84)	37.4 (3.66)	36.5 (3.15)*	34.1 (4.03)*	38.15 (6.02)	37.65 (5.10)	39 (6.64)	35.1 (6.60)
Height DJ (cm)	29.6 (6.44)	34.15 (7.11)	30.05 (6.42)	27.4 (6.49)	29.75 (8.18)	30.3 (5.54)	26 (10.45)	27.7 (4.77)
Relative Power SJ (W·kg-1)	47.75 (5.33)	45.9 (3.71)	45.05 (5.61)	43.91 (3.68)	45.75 (3.71)	45.9 (3.55)	44.95 (6.31)	44.4 (4.57)
Relative Power CMJ (W·kg-1)	49.9 (4.80)	48.25 (3.39)	47.75 (2.51)*	45.4 (3.32)*	48.7 (4.93)	48.5 (4.30)	49.7 (6.01)	46.22 (5.50)
Relative Power DJ (W·kg-1)	42.14 (5.93)	45.51 (5.48)	41.75 (4.80)	40.23 (4.91)	41.8 (6.84)	42.25 (4.59)	38.55 (9.67)	40.15 (3.75)
1RM (kg)	143 (29.45)	103 (10.42)	128 (26.82)	102 (10.94)	140 (29.32)	100 (10.73)	142 (32.11)	99 (11.40)
Relative Strength (u.a.)	1.78 (0.24)	1.35 (0.08)	1.68 (0.24)	1.31 (0.09)	1.73 (0.24)	1.32 (0.07)	1.72 (0.27)	1.32 (0.10)
VO$_2$max (ml·kg-1·min-^{1})	44.46 (2.31)	46.09 (2.44)	42.29 (2.35)	43.75 (3.05)*	43.63 (1.76)	43.88 (3.37)	43.36 (1.72)	43.99 (3.94)

Data are presented as mean ± SD.

Note: SJ, Squat Jump; CMJ, Counter Movement Jump; DJ, Drop Jump; RM, maximum repetition; a.u., arbitrary units; LT, lower-trained group; HT, higher-trained group; *significant differences between moments compared to pre-test.

6 DISCUSSION

The aim of this study was to verify acute MD and recovery behavior after one HIFT session through biochemical markers and performance indicators (according to participants divided in two different groups based on their fitness status). To evaluate fitness status and classify them to higher-trained (HT) or lower-trained (LT) groups, lower limbs relative strength (1.82 ± 0.24 a.u. and 1.35 ± 0.08 a.u., respectively) was accessed through the squat exercise and training history (physically active for at least one year). To analyze exercise-induced MD and acute recovery process, indirect biochemical markers for MD (CK and LDH) and performance (strength, power and aerobic capacity) were investigated pre-exercise, immediately after, 24h and 48h after HIFT bout. The results for CK and LDH revealed that MD was detected for both groups, with a suggestion for faster homeostasis restoration and biochemical markers returning to basal levels for HT. Performance data also revealed MD for both groups, once lower-limb power was impaired after HIFT. Post session, LT group experienced a decrease in capacity during the aerobic capacity test due to fatigue related to HIFT session. Performance data showed that pre-test values and after 48h were closer for the HT group, suggesting a faster recovery. Therefore, the hypothesis was partially accepted, once we could not demonstrate less MD to HT, but it was possible to imply a faster recovery for this group.

A novel finding is that CK or LDH did not change across the time between LT and HT groups. Also, HT group presented higher CK activity pre-HIFT when compared with LT group, and the CK activity was significantly increased after 24h when compared to post-HIFT workout on HT group. Our hypothesis was not confirmed since the HT group demonstrated higher values of MD after the HIFT workout. This is not the first study to observe physiological impairments pre-HIFT workout in higher trained participants. Perciavalle *et al.* (2016) investigated the lactate responses in professional CrossFit® female athletes performing a HIFT workout. The values of lactate pre-HIFT workout were considered higher than those which normally occur at rest (4.5 mmol). Timón *et al.* (2019) found that two days of HIFT workout in trained male participants increased the CK values, returning its values to basal after 48h of HIFT workouts. CK which represents muscle damage and metabolism may provide information about training intensity. Therefore, the results of the present study suggest that muscle damage occurs after a single HIFT-workout in trained participants, probably due to its already demonstrated ability to perform exercises at greater intensities (Meier; Sietmann; Schmidt, 2022).

Although the CK values were higher in the HT group, it did not affect the participants' ability to recover the lower limbs performance. The HT and LT groups decreased their lower limbs power performance post-HIFT workout, but it was reestablished after 24h after the workout. Tibana *et al*. (2016) reported that two days of HIFT workout impaired pro- and anti-inflammatory cytokines and Osteoprotegerin without impairments in lower limbs muscle power in experienced HIFT workout male participants. These results corroborate with previous studies of our research group, which recently demonstrated that two days of simulated competition did not impair anaerobic power or fatigue in HIFT athletes (Zecchin *et al*., 2022). Timón *et al*. (2019) investigated two days of HIFT workouts (two workouts containing weightlifting, metabolic and gymnastic exercises) on biochemical parameters and physical performance (plank test) in trained HIFT participants (VO_2max: 47.8 ± 3.6 $ml.kg.min^{-1}$, 1RM power clean: 93.2 ± 7.6kg). Both, physical performance and biochemical parameters such as blood glucose, hepatic transaminases, and CK were impaired for 24h after the workouts completed, returning to basal level after 48 the workouts were finished.

Regarding VO_2max, only the LT group decreased the VO_2max post-HIFT workout, reestablishing its values after 24h HIFT workout. Interestingly, there were no differences in pre-VO_2max test between groups, however, the LT group decreased their VO_2max post-HIFT workout. Here, the HT group did not decrease their VO_2max post-HIFT workout. Studies evaluating participants from different training levels revealed that in part, the movement economy (ME) displays an important role on training and racing performance (Barnes; Kilding, 2015). In brief, factors such as metabolic efficiency, cardiorespiratory efficiency, training experience, biomechanical efficiency, and neuromuscular efficiency are capable of determining the most ME athletes. Although we did not evaluate the ME, it is expected that the HT group would have better ME than LT group.

It is important to emphasize that both groups were previously trained (Mchugh *et al.*, 1999), which means that there were no untrained individuals, therefore all of them were protected (perhaps not fully, but partially) from MD by RBE phenomenon. This theory explains the reasons why well-trained individuals, presenting higher performance levels, probably suffer a lower exercise-induced MD. Cellular, neural, and mechanical (connective tissue) adaptations justify the protection-effect acquired, once the system becomes prepared to receive repetitive stimulus. Other mechanisms such as adaptations in muscle contraction cycle and inflammatory responses after MD can be related to RBE (Mchugh *et al.*, 1999). Knowing that, it was expected that no extraordinary differences would happen between groups.

Enzymatic activity of CK and LDH are a well-known tools to access MD, since they are located inside the muscle cell and incapable of crossing the membrane, indicating that their extravasation into the bloodstream is due to MD (Aquino *et al.*, 2016 and Brancaccio; Maffulli; Limongelli, 2007). No statistical differences were found for LDH activities at the same moment between groups, but kinetics of removal during recovery shows inferiority of LT in comparison to HT, especially when observing the behavior of the moment 24h to 48h after HIFT, in which LT group increased activity to a point of exceeding HT value, distancing it from baseline levels. For CK activity, the LT decreased the CK activity immediately after the exercise while HT increased the same component immediately after the exercise. It suggests the presence of MD for the HT group, because despite trying to equalize training load by RPE and 70% of 1RM, HT group performed more repetitions within the periods of effort stipulated (data not shown). Therefore, it is hypothesized that they trained at a higher intensity, since exercise effort is better tolerated by them. Although it was not recorded to be posteriorly analyzed from a statistical view, it could be a possible explanation (and also a limitation of the study) why the HT group probably experienced a higher MD based on CK activity. Even showing a higher MD, kinetics behavior of CK for the HT group over the period of this study (48h) suggests that they tend to restore homeostasis (*i.e.*, recovery) before the LT.

In contrast, accessing MD, fatigue and recovery related to training status from perspective of performance, it is possible to suggest that HIFT session induced MD for both groups regardless the training status, seen variables such as height and relative power on CMJ at post-session moments showed a significant difference when compared to groups separately. Only LT reduced the oxygen consumption right after HIFT bout ($p < 0.05$). The aerobic capacity test was the last one to be performed and a decrease in VO_2max does not mean a drop in oxygen consumption per se, but a loss in the ability to execute the movement of running on the treadmill. Performance tests presented that, immediately after training session, there was a considerable performance decrease to individuals about variables mentioned before. However, it is important to notice that the HT group at the moments post, 24h and 48h after session performed values of maximal load very close to pre-session values. The opposite occurred to the LT group, which showed a decrease at the maximal load test at post-session moment and, on the average, continued decreasing performance 24 and 48 hours after training. Although speculative, it suggests that relative strength levels interfere directly on individuals exercise-induced MD and recovery capacity after HIFT session accomplishment. (Johnston; Gabbett; Jenkins, 2015) found similar results to the present study, they concluded, after examining the influence of physical qualities on markers of fatigue and MD post-match in rugby players, that individuals with well-developed lower limbs strength experienced less MD and fatigue, despite these players having greater internal and external match loads.

The MD and fatigue process is widely investigated due to the requirement of not only avoiding overtraining but allowing better enhancement in performance. In the eagerness to accumulate training sessions to improve performance, practitioners must organize the sessions in a way that allows an adequate recovery and, moreover, an overcompensation of performance. Thinking about it, several studies stipulate the number of sessions from 2 to 3 times a week (for resistance-based exercises), considering that at least 48 hours are necessary to restore organism from MD and fatigue to basal levels (Schlegel, 2020).· Doeven *et al.*, (2018), in a study about post-match recovery of physical performance and biochemical markers in team ball sports, affirm that for biochemical markers as CK 72 hours are necessary to ascertain full recovery, while in addition it is determined that CMJ height recover faster than CK. Physical performance recovery takes up to ≥ 48 hours after regular training. In agreement with the last statement Magalhaes *et al.* (2010) analyzed MD and neuromuscular function throughout 72 hours of recovery after an intermittent running test and a soccer match. They reported decrements in force-generating capability that lasted 24-48 hours after exercise when soccer players were submitted to the intermittent running test. Over again, another study from (Ascensão *et al.*, 2008), that aimed to analyze the effect of a competitive soccer match on plasma levels of oxidative stress and MD markers, and to relate these findings with lower limb functional data revealed that lower limb strength and sprint ability were lower and CK levels were higher than baseline throughout 72 hours recovery period in soccer players.

There is a lack of studies comparing training status among well-trained individuals. Mostly, literature approaches trained *vs.* untrained population. Besides that, traditional HIIT (endurance-based exercise) has advantages in the research field due to its popularity, which does not happen to HIIT resistance-based training or HIFT (except for the growing enthusiasm to CrossFit®). Moreover, there are many aspects that influence recovery which were not investigated. Dietary intake was not recorded after HIFT session, impairing evaluation about adequate availability of carbohydrates and proteins that affect recovery, *e.g*. Also, performance in the post-session tests may have been influenced not only by MD, but by acute depletion of substrates needed to ensure performance (*e.g.*, glycogen and phosphate stores) (Johnston; Gabbett; Jenkins, 2015). Furthermore, even with biochemical markers and performance data revealing the presence of MD and fatigue or absence of complete recovery, it does not mean that accomplishing a HIFT session will culminate in impairments for organism and performance. Our findings reveal the acute effects of HIFT on MD and recovery regarding training status, additional research should investigate the chronic effect within different periodizations.

This manuscript is not free of limitations, among them we can mention: i) Many aspects that influence recovery were not investigated, ii) the absence of a control group, iii), diet and resting control of the participants were not performed in the periods before the collections. Our findings reveal the acute effects of HIFT-workout on MD and recovery regarding training status, additional research should be conducted aiming to understand the chronic effects of HIFT-workout on these variables. Besides that, this study strengthens the evidence that trained subjects are able to exercise more for the same relative intensity than less trained subjects. Additionally, the time required for the restoration of their biological functions seems to be shorter.

7 CONCLUSION

The HIFT workout negatively affected the power performance in both groups but its value was recovered post-24h and, VO_2max showed impairment post-HIFT workout only in the LT group. Finally, the biochemical CK activity was altered pre-HIFT workout in HT group, which may contribute to the increased activity of CK 24h-post HIFT in HT group. Strength and conditioning trainers should be aware about the negative impacts on metabolic and performance level in trained HIFT participants. The long-term effects of the HIFT workout in the performance and physiological variables has to be proven in future studies.

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