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**ACUTE EFFECTS OF AN UPPER BODY REPEATED SPRINT PROTOCOL IN HYPOXIA
INDUCED BY VOLUNTARY HYPOVENTILATION**

Dissertação elaborada com vista à obtenção do Grau de Mestre em Treino de Alto
Rendimento

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Abstract

Voluntary hypoventilation at low lung volume (VHL) consists of breath holding with partly empty lungs after an exhalation, leading to the functional residual capacity (Woorons, 2014). When combining with exercise, VHL induces severe arterial oxygen desaturation, as well as muscular and cerebral deoxygenation, causing both an hypoxic and hypercapnic effect. When combined with repeated sprint exercise (RSE), VHL has showed great improvements in repeated sprint ability (RSA) attributed to enhanced glycolytic utilization and buffer capacity. Previous studies have analyzed a wide range of modalities, running, cycling, and swimming. Given that Brazilian Jiu Jitsu (BJJ) could benefit with this kind of training we propose to investigate metabolic, muscle oxygenation and performance acute effects of an upper body repeated sprint protocol with VHL.

Eighteen male well trained BJJ athletes (mean \pm SD; age 32 ± 7.3 years; body mass 73.75 ± 11.1 kg;) of national and international level participated in this study. This was an experimental study with a randomized crossover design. Participants performed three sessions, a familiarization session and two moments of evaluation, consisting of a repeated sprint protocol with normal breathing (RSN) and the other using the same repeated sprint protocol with VHL (RSH-VHL) in an arm cycle ergometer. Peak power output (PPO), mean power output (MPO) and total work (TW) were measured, also repeated sprint ability decrement score (RSA_{decs}) was calculated. Arterial oxygen saturation (SpO_2), heart rate (HR), gas exchange and muscle concentrations of oxyhaemoglobin/myoglobin (O_2Hb) and deoxyhaemoglobin/myoglobin (HHb) were continuously recorded throughout exercise. Blood lactate concentration ($[La]$) was measured at the end of the first (S1) and second set (S2). Bench press throw peak power (BP_{PPO}) and isometric hand grip strength (HG_{iso}) were recorded before and after the repeated sprint protocol in order to assess differences.

RSN MPO (162 ± 3.83 SE) was greater than MPO in RSH-VHL (156 ± 4.0 SE), $p < 0.01$. On the other hand, RSA_{decs} and bench press throw peak power drop after the repeated sprint protocol were not impacted by VHL. A significant SpO_2 drop was observed at the end of the second set in RSH-VHL (97.62 ± 2.60) when compared with RSN (98.71 ± 1.86), $p = 0.03$ but there were no significant changes in muscle deoxygenation. Blood lactate concentration was lower in RSH-VHL (8.04 ± 3.00) when compared with RSN (10.42 ± 2.55), $p < 0.01$. Finally, RSH-VHL (31.12 ± 1.11 SE) induced greater $\dot{V}O_2$ when compared with the same exercise in normal breathing conditions (29.65 ± 0.87 SE), $p = 0.03$.

In conclusion this study revealed that although statistically significant the reduction of the arterial oxygen saturation with the utilization of ventilatory hypoventilation with low lung volume may not have been sufficient

to induce significant levels of hypoxia. MPO and PPO were significantly affected by RSH-VHL but there was not a significant decrease in total work or RSA_{decs} . Blood lactate concentration and respiratory exchange ratio were lower in RSH-VHL when compared with RSN and $\dot{V}O_2$ was greater.

Keywords: Voluntary hypoventilation with low lung volume, hypoxia, repeated sprinting, repeated sprinting in hypoxia, arm cycle ergometer, Brazilian Jiu-Jitsu.

Resumo

A hipoventilação de baixo volume pulmonar (VHL), consiste em sustentar a respiração com pulmões parcialmente vazios após uma exalação forçada, conduzindo à capacidade residual funcional (Woorons, 2014). Quando combinado com exercício, o VHL induz a dessaturação severa do oxigênio arterial, bem como a desoxigenação muscular e cerebral, causando um efeito hipóxico e hipercápnico. Quando combinado com o exercício de sprints repetidos, o VHL tem mostrado grandes melhorias na capacidade de sprint repetida atribuída a uma melhor utilização glicolítica e capacidade tampão. Tem sido usado numa vasta gama de modalidades, como corrida, ciclismo e natação. Uma vez que o Jiu Jitsu Brasileiro (BJJ) poderia beneficiar com este tipo de treino, propomo-nos investigar os efeitos agudos metabólicos e musculares de um protocolo de sprint repetidos na parte superior do corpo com VHL.

Dezoito atletas de BJJ masculinos bem treinados (média \pm SD; idade 32 ± 7.3 anos; massa corporal 73.75 ± 11.1 kg;) de nível nacional e internacional participaram neste estudo. Este foi um estudo experimental com um design de crossover aleatório. Os participantes realizaram três sessões, uma sessão de familiarização e dois momentos de avaliação, que consistiram em realizar o protocolo de sprints repetidos com a respiração normal (RSN) e outra utilizando o mesmo protocolo de sprints repetidos com VHL num ciclo-ergómetro de braços. A potência máxima (PPO), a potência média (MPO) e o trabalho total (TW) foram medidos, também foi calculado o índice de fadiga em sprints repetidos (RSA_{decs}). A saturação de oxigênio arterial (SpO_2), a frequência cardíaca (HR), a troca de gases as concentrações musculares de oxi-hemoglobina (O_2Hb) e desoxihemoglobina /mioglobina (HHb) foram continuamente registados durante todo o exercício. A concentração de lactato sanguíneo ([La]) foi medida no final da primeira série e na segunda série. A potência máxima de lançamento da barra de supino (BP_{PPO}) e força de prensão manual (HG_{iso}) foram registadas antes e depois do protocolo de sprints repetidos, a fim de avaliar as diferenças.

RSN MPO ($162 \pm 3,83$ SE) foi maior que MPO em RSH-VHL (156 ± 4.0 SE), $p < 0.01$. Por outro lado, a RSA_{decs} e a potência máxima do lançamento da barra de supino após o protocolo de sprints repetidos não foram impactados pela VHL. Uma queda significativa do SpO_2 foi observada no final da segunda série em RSH-VHL (97.62 ± 2.60) quando comparado com RSN (98.71 ± 1.86), $p = 0.03$, mas não houve alterações significativas na desoxigenação muscular. A concentração de lactato sanguíneo foi menor em RSH-VHL (8.04 ± 3.00) quando comparada com RSN (10.42 ± 2.55), $p < 0.01$. Por fim, o RSH-VHL (31.12 ± 1.11 SE) induziu maior $\dot{V}O_2$ quando comparado com o mesmo exercício em condições normais de respiração (29.65 ± 0.87 SE), $p = 0.03$.

Em modo de conclusão, este estudo revelou que, embora estatisticamente significativo, a redução da saturação do oxigênio arterial com a utilização da hipoventilação com baixo volume pulmonar pode não ter sido suficiente para induzir níveis significativos de hipoxia. A MPO e a PPO foram significativamente afetadas pela RSH-VHL, mas não houve uma diminuição significativa do trabalho total ou do RSAdec. A concentração de lactato sanguíneo e quociente respiratório foram mais baixos em RSH-VHL quando comparados com RSN e $\dot{V}O_2$ foi maior.

Palavras-chave: Hipoventilação voluntária com baixo volume pulmonar, hipoxia, sprint repetido, sprints repetidos em hipoxia, ciclo-ergómetro de braços, jiu-jitsu brasileiro.

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List of Abbreviations

- [La] – Blood lactate concentration
- [La]_{max} – Maximal blood lactate concentration
- ANOVA – Analysis of variance
- ATP – Adenosine triphosphate
- BFR – Blood flow restriction
- BJJ – Brazilian Jiu-Jitsu
- Bpm – Beats per minute
- BP_{PPO} – Bench press throw peak power
- CO₂ – Carbon dioxide
- HG_{iso} – Isometric hand grip strength
- HHb – Deoxyhaemoglobin
- HIIT – High intensity interval training
- HIT – High intensity training
- HR – Heart rate
- HR_{mean} – Mean heart rate
- IHE – Intermittent hypoxic exposure
- IHIT – Interval hypoxic training
- IHT – Continuous hypoxic training
- IPC – Ischemic pre-conditioning
- LHTH – Living high training high
- LHTL – Living high training low
- LLTH – Living low training high
- MPO – Mean power output
- NIRS – Near infrared spectroscopy
- O₂ - Oxygen
- O₂Hb – Oxyhaemoglobin
- PAR-Q & YOU – Physical activity readiness questionnaire
- P_{CO2} – Partial pressure of carbon dioxide

PCr – Phosphocreatine

PPO – Peak power output

RER – Rate of exchange ratio

RPE – Rating of perceived exertion

RSA – Repeated sprint ability

RSA_{decs} – Repeated sprint ability percentage decrement score

RSE – Repeated sprint exercise

RSH – Repeated sprint in hypoxia

RSH – Repeated sprint in hypoxia

RSH-VHL – Repeated sprint in hypoxia with voluntary hypoventilation at a low lung volume

RSN – Repeated sprint with normal breathing

RTH – Resistance training in hypoxia

S1 – First set

S2 – Second set

SD – Standard deviation

SE – Standard error

SIH – Sprint interval training in hypoxia

SpO₂ – Arterial oxygen saturation

tHb - Total haemoglobin

TOI - Tissue saturation index

TW – Total work

\dot{V}_E – Volume of expired air

VHL – Hypoventilation with low lung volume

$\dot{V}O_2$ – Oxygen consumption

$\dot{V}O_{2max}$ – Maximal oxygen consumption

$\dot{V}O_{2peak}$ – Peak oxygen consumption

1. Introduction

Voluntary hypoventilation at low lung volume (VHL) consists of breath holding with partly empty lungs after an exhalation, leading to the functional residual capacity, which is a bit less than 50% of total lung capacity (Woorons, 2014). When combining with exercise, VHL induces severe arterial oxygen desaturation, leading to muscular and cerebral deoxygenation (Kume et al., 2013, 2016; Woorons et al., 2007, 2010, 2011), causing both an hypoxic and hypercapnic effect (Woorons, 2014; Woorons et al., 2010). Higher [La] with VHL was observed (Kume et al., 2013, 2016), along with lower muscle oxygenation (Kume et al., 2013, 2016), suggesting a greater energy supply from the anaerobic glycolysis. With an intermittent and brief exposure to hypoxia it is considered to be an intermittent hypoxic method within “Living Low-Train High” hypoxic model (LLTH).

In combination with repeated sprint exercise (RSE), VHL has showed greater oxygen uptake (Woorons & Dupuy, et al., 2019; Woorons et al., 2017) and lower [La] at the end of exercise and the authors attributed these results to a better lactate clearance (Woorons et al., 2017; Woorons, Millet, et al., 2019). Additionally it has been shown that there is an improvement in RSA after RSE training in combination with VHL (RSH-VHL) in swimming (Trincat et al., 2017; Woorons et al., 2016), cycling (Woorons et al., 2020; Woorons, Millet, et al., 2019) and running (Fornasier-Santos et al., 2018; Lapointe et al., 2020), making the VHL approach a recognized and a potentially useful training method for a wide range of sporting activities (Millet et al., 2019). Despite the repeatedly improvements reported of RSH-VHL on RSA, the underlying mechanisms that justify such adaptations, are yet to be fully understood and explained (Girard et al., 2017, 2020; Millet et al., 2019).

Given that glycolytic pathway may be potentially improved with RSH-VHL, activities that predominantly use these energy pathways are more likely to benefit from the use of this method.

Included in this category is Brazilian Jiu-Jitsu (BJJ). BJJ is a grappling combat sport, and it is considered intermittent in terms of its energetic demands. This intermittency of the effort is characterised by short bouts of high-intensity and explosive movements interspersed by moderate or low-intensity periods (Andreato et al., 2017). Although predominantly aerobic, post-match samples of lactate ($\approx 10 \text{ mmol.L}^{-1}$) suggest a moderate to high glycolytic activation (Andreato et al., 2016).

It is why that high intensity interval training (HIIT) is suggested to be used in BJJ strength and conditioning programs, to cope with the high-intensity intermittent effort pattern and specificity of physiological demands (Franchini, Cormack, et al., 2018; Vasconcelos et al., 2020). We can distinguish four main types of HIIT protocols: HIIT using long-duration intervals; HIIT using short-duration intervals, sprint interval training and

repeated sprint training or repeated sprint exercise (RSE) (Buchheit & Laursen, 2013; Girard et al., 2011). Where repeated sprint ability (RSA) translates the capacity to reproduce sprint bouts throughout the time or competition (Girard et al., 2011). RSE or some kind of HIIT have been used extensively, but just recently tested and developed in hypoxic environments with an increased interest (Girard et al., 2011; Millet et al., 2019).

All the studies that have used RSH-VHL, applied those protocols in running, cycling, or swimming. From a combat sports perspective, more specifically those who use *gi* (e.g. Brazilian Jiu-Jitsu and Judo), it has been reported that the upper limbs are more likely to have local fatigue between matches and throughout the competition, when compared with lower limbs, furthermore this is an indicator that distinguishes experienced and novice athletes (Andreato et al., 2017; Bonitch-Góngora et al., 2012; Franchini, Schwartz, et al., 2018; Kons et al., 2018).

A recent study from Willis, et al. (2019), found that during an RSE protocol in an arm ergometer, with induced systemic hypoxia and with blood flow restriction, arms were more sensitive to local deoxygenation than legs. Furthermore Beard et al. (2019), found improvements in RSA power outputs, after an upper limb repeated sprint in hypoxia (RSH) protocol, when compared with the same training protocol in normoxia. Such results suggest a potential use of the VHL technique in combination with upper limbs exercise.

Therefore, the purpose of this study is to compare the metabolic, muscle oxygenation and performance acute effects of upper body RSH-VHL protocol with RSN, in Brazilian Jiu-Jitsu fighters. Additionally, we propose to analyse the impact of RSH-VHL protocol on explosive movements. We suggest that acute effects will be significantly different between both protocols, moreover it will be possible to see changes in both central and local responses when comparing RSH-VHL and RSN. We expect to see a decrement in power output which magnitude may be greater in the RSH-VHL protocol when compared with RSN. We also speculate that RSH-VHL may lead to higher arterial and muscle deoxygenation throughout the exercise. Finally, we hypothesize that RSH-VHL may induce an increased blood lactate concentration, when compared with RSN.

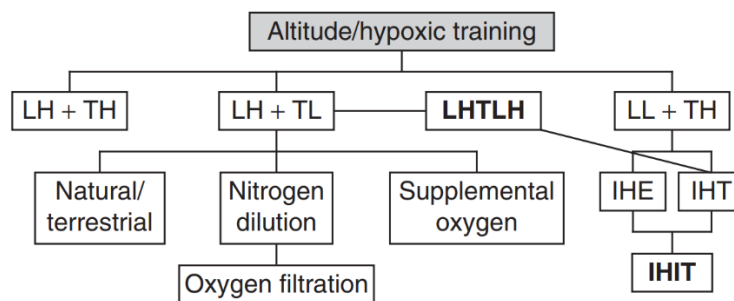
From what we know, this will be the first study to apply and analyse the acute effects of VHL technique in combination with a RSE protocol in an upper limb exercise.

2. Literature Review

2.1. Hypoxic Training Approaches

Since 1968 Olympics the outstanding results (attributed to altitude acclimatization) of East Africans long distance runners, stimulated Western lowlander athletes to routinely implement altitude training in their preparation to major competitions (de Smet, 2019). Although the underlying mechanisms about the effects and the application of hypoxic training are still widely debated, it is considered that altitude training may lead to enhanced haematological capacity which results in an improved maximal oxygen consumption that may benefit endurance aerobic sports (Millet et al., 2010, 2019). This may not be the only factor involved in the performance improvement, other central, such as ventilatory, haemodynamic and neural adaptations may take place as well as peripheral such as muscle buffering capacity or economy (Millet et al., 2010).

Figure 1 – Different altitude training models, Millet et al. (2010)



It is no surprise that the early 1970s marked the starting point of the scientific investigation on the effectiveness of altitude training (Millet et al., 2019). At this time, several altitude training centres around the world were developed for the exclusive purpose of training and living in high altitude, the so-called Live High – Train High model (LHTH) (Millet et al., 2019).

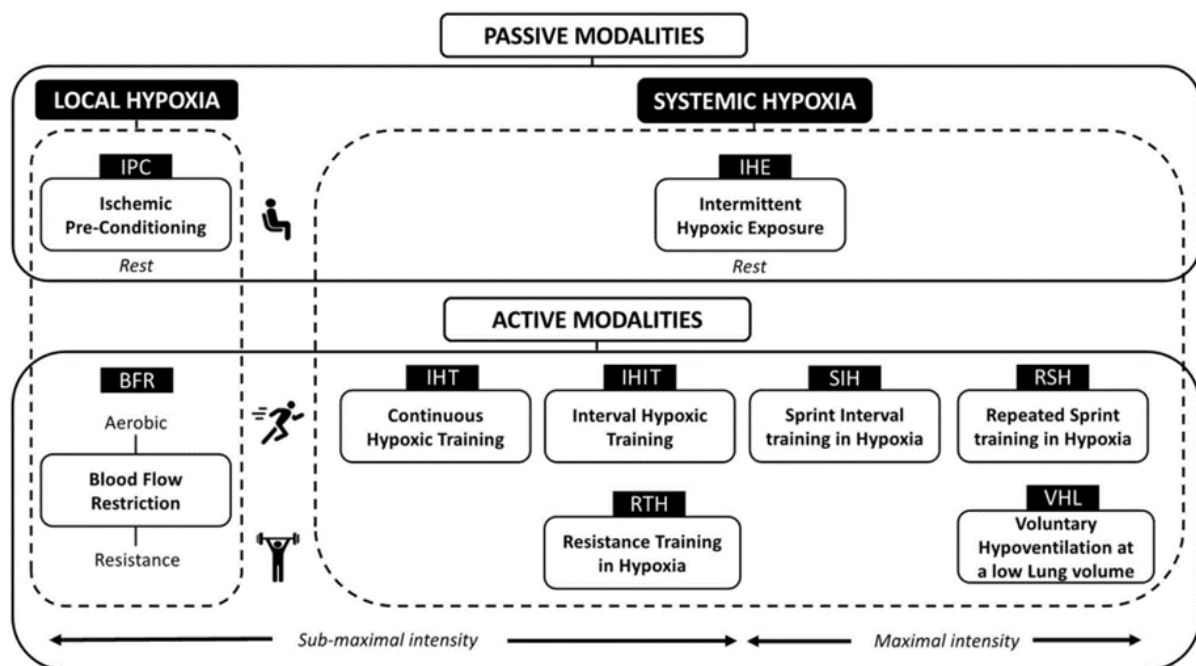
In 1997, Levine & Stray-Gundersen showed high-intensity endurance performance (5000 m run, ≈16-18 min) improvements following “living high – training low” (LHTL), but not after LHTH. Since then, the LHTL concept is the preferred altitude training strategy over LHTH (de Smet, 2019), in recent times, with minimal costs and travel constraints for athletes, the “Live Low-Train High” (LLTH) approach is becoming an important intervention for modern sport (Girard et al., 2020; Millet et al., 2019). See figure 1.

2.2. Living Low – Training High hypoxic approach

LLTH methods are characterised by brief exposures to hypoxia while resting, usually less than 2 hours, or while training, typically between 2-5 times per week (Girard et al., 2020).

The popularity of the LLTH model of altitude training is also due to the fact that athletes' usual daily routine is not much affected, allowing them to keep their regular lifestyle in their home environment (Girard et al., 2020). Additionally, sleep quality or recovery is well preserved since the athletes do not sleep in hypoxia (Girard et al., 2020). The potential benefits of LLTH are highly individual, as well as the emphasis of their training also differs if it is predominantly aerobic or anaerobic (Girard et al., 2020).

Figure 2 - Living Low - Training High modalities and methods, adapted from Girard et al. (2020)



Included in this type of hypoxic training, a myriad of new methods have emerged. Passive modalities include intermittent hypoxic exposure at rest (IHE) and ischemic pre-conditioning (IPC). IHE implies the use of short periods (3–6 min) of relatively severe levels of hypoxia ($F_{iO_2} = 0.15-0.09$; 2800–5500 m simulated altitude) combined with normoxic episodes of similar duration (Girard et al., 2020). If there are some doubts over IHE effectiveness, it was observed a 1–5% gain in time-trial performance and aerobic capacity after IPC (Girard et al., 2020). IPC consists in causing tissue ischemia via circumferential compression of limb(s) followed by reperfusion in a repeated/cyclic approach (Girard et al., 2020).

However, possible improvements in exercise capacity resulting from LLTH interventions depend on the type and intensity of the workout considered, through local hypoxia [blood flow restriction (BFR)] or systemic hypoxia exposure at submaximal intensities [continuous hypoxic training (IHT), interval hypoxic training (IHIT)] or resistance training in hypoxia (RTH) or maximal intensities [sprint interval training in hypoxia (SIH) or RSH] (Girard et al., 2020).

BFR involves limiting arterial blood supply and venous return, usually at the limbs, while exercising (Girard et al., 2020). BFR has been applied during both, resistance exercise and aerobic exercise, and also passively without exercise. Other applications in combination with BFR involves non-traditional exercise modalities, such as whole-body vibration techniques and neuromuscular electrical stimulation (Patterson et al., 2019). Willis, et al., (2019) have implemented it with repeated sprint exercise, leading to muscle excitation–contraction coupling probably due to an immediate muscle deoxygenation and impairment in metabolite removal ability during exercise, that have impaired RSA.

Continuous hypoxic training, as its name implies, relates to continuous sub-maximal training sessions under hypoxic conditions, it is usually utilized to improve endurance-based performance although given some practical limitations the results suggest that there are no changes when compared with exercising in normoxia, the only studies that found some benefit included some kind of high intensity interval training (Girard et al., 2020; McLean et al., 2014). Regarding IHIT the majority of well-controlled studies report no additional benefit of the hypoxic stimulus, although it has been reported to evoke cellular responses via hypoxia inducible factors (HIFs) (Faiss et al., 2013; McLean et al., 2014). Recently, following the first BFR studies, resistance training has been researched in hypoxic conditions because it is thought to potentiate some of its effects, although methodological flaws in RTH studies to date prevent any definitive conclusions about the effectiveness of LLTH for enhancing performance gains (McLean et al., 2014).

It is known that RSN increases the capacity for anaerobic energy production and enhance metabolic acidosis tolerance due to increased muscle buffer capacity among other factors (Girard et al., 2011; Hargreaves & Spriet, 2020). However, results from studies conducted in the last decade suggest that RSH leads to superior RSA improvements (i.e., faster mean sprint times or higher power outputs associated with a better resistance to fatigue during a repeated-sprint test) when compared with RSN (Millet et al., 2019). The potential mechanisms include transcriptional factors involved in oxygen-signalling and oxygen-carrying capacity and mitochondrial metabolism enzymes, improved behaviour of fast-twitch fibers via compensatory vasodilatation, improved vascular relaxation

and greater microvascular oxygen delivery as well as faster rate of phosphocreatine resynthesis (Millet et al., 2019).

Briefly RSH consists in the repetition of short (<30-s) “all-out” sprints with incomplete recoveries (<60-s) in hypoxia and include exercise-to-rest ratios typically lower than real context repeated-sprint ability tests/in-match, in order to induce a metabolic stress increase (Girard et al., 2020).

However, traveling to altitude training centres (hypobaric hypoxia) is not always viable (i.e., travel time, athlete engagement, expenses) for all but the really top athletes or teams. Additionally, access to simulated altitude facilities (normobaric hypoxia) for permanent residence or training is costly and logistically difficult for a large number of athletes (Girard et al., 2020).

2.3. Hypoventilation with low lung volume

When combined with exercise VHL, can induce reductions in SpO₂ up to about ~75% (non-presented data) which may correspond to the same value obtained at approximately 5500m (Lorente-Aznar et al., 2016).

Previous works comparing exercise at submaximal intensities showed an increased cardiac work through a greater heart rate (HR), stroke volume, cardiac output, mean arterial pressure and sympathetic modulation when combined with VHL (Woorons et al., 2007, 2011). Higher [La] with VHL was observed (Kume et al., 2013, 2016), along with lower muscle oxygenation (Kume et al., 2013, 2016), suggesting a greater energy supply from the anaerobic glycolysis. Furthermore, Kume et al. (2016), reported an electrical muscle activity increase after an RSH-VHL protocol, due to muscle deoxygenation and accumulation of muscle metabolites or greater recruitment of type II fibres, potentially due to hypercapnic effects.

When combining with RSE, greater oxygen uptake was reported (Woorons & Dupuy, et al., 2019; Woorons et al., 2017) and lower blood lactate concentration ([La]) at the end of RSH-VHL protocol, the authors credited this to a better lactate clearance (Woorons et al., 2017; Woorons, Millet, et al., 2019).

Moreover, after 10 sessions of training at submaximal intensities in combination with VHL, it was observed a reduced exercise-induced blood acidosis that was attributed by the authors to an improvement in muscle buffer capacity and therefore to positive improvements in anaerobic performance (Woorons et al., 2008).

Additionally it has been shown that there is an improvement in RSA after RSE in combination with VHL (RSH-VHL) in swimming (Trincat et al., 2017; Woorons et al., 2016), cycling (Woorons et al., 2020; Woorons, Millet, et al., 2019) and running (Fornasier-Santos et al., 2018; Lapointe et al., 2020), making the VHL approach a recognized and a potentially useful training method for a wide range of sporting activities (Millet et al., 2019).

Trincat et al. (2017), reported an increase in the number of sprints after RSH-VHL in swimming (7.1 ± 2.1 vs. 9.6 ± 2.5 ; $p < 0.01$). This increase, in the number of sprints was also observed in running (+64%, $p < 0.01$) after RSH-VHL (Fornasier-Santos et al., 2018). In this study not only RSH-VHL group made more sprints but also their mean velocity was greater throughout the entire test protocol. Moreover, whenever it was possible to measure power output, mean power output (MPO) post RSH-VHL ($p < 0.01$) was also greater (Woorons et al., 2020).

Among the main physiological effects it was observed a significative arterial oxygen saturation (SpO_2) drop (Fornasier-Santos et al., 2018; Trincat et al., 2017; Woorons et al., 2016, 2020; Woorons, Millet, et al., 2019), a $[La]$ increase after RSH-VHL or supramaximal intensity training, which can be translated has an improved lactate tolerance (Trincat et al., 2017; Woorons et al., 2016; Woorons, Millet, et al., 2019), also an increased P_{CO_2} and a greater O_2 uptake was reported although it was not verified in all studies (Woorons, Millet, et al., 2019), There was no significative changes in the breathing frequency and tidal volume (Woorons et al., 2016; Woorons, Millet, et al., 2019), also there has been no difference in HR after RSH-VHL training (Fornasier-Santos et al., 2018; Trincat et al., 2017; Woorons et al., 2016, 2020; Woorons, Millet, et al., 2019). The last observations suggest an overall improved cardiorespiratory efficacy after RSH-VHL training, this was verified when comparing other parameters such as oxygen pulse ($\dot{V}O_2/HR$) where these values were higher (Woorons, Millet, et al., 2019). Such fact would make the authors claim that in this last study, RSA improvement after RSH-VHL, unlike RSH, could be mainly due to central rather than muscular adaptations.

However, Woorons, Millet, et al. (2019), reported a 6% gain in a post-Wingate test emphasising the anaerobic contribution and positive change after RSH-VHL training. Moreover, in a recent study Woorons et al. (2020), reported that despite the increase in RSA, RSH-VHL training in cycling did not increased performance or $[La]_{max}$ in a 200m maximal run trial, suggesting that the physiological adaptations leading to an improved anaerobic glycolysis after HIT with VHL, were highly exercise specific and therefore highly dependent on local adaptations. Accentuating the fact that even if there are central adaptations, RSH-VHL still imply local adaptations that cannot be ignored.

2.4. Brazilian Jiu Jitsu characteristics and physiological demands

Brazilian Jiu-Jitsu (BJJ) is a grappling combat sport and it is considered intermittent in terms of its energetic demands. This intermittency of the effort is characterised by short bouts of high-intensity and explosive movements interspersed by moderate or low-intensity periods (Andreato et al., 2017). Time motion analysis report an effort:pause ratio of 6:1 to 13:1, with efforts duration between 8 and 290 seconds and pauses from 5 to 44 seconds (Andreato et al., 2016). Low-intensity efforts last longer and have a higher contribution during each match than those with high intensity, demonstrating an aerobic metabolism predominance however, post-match samples of lactate (10 mmol.L^{-1}) suggest a moderate to high glycolytic activation (Andreato et al., 2016).

The competition schedule often demands the athletes to perform six matches during a day of competition and this number may increase in the open-class competition, which is carried out parallel to the competition by weight class (Andreato et al., 2017). Such fact elicits a high level of conditioning to support optimal levels of performance along the match time and throughout the competition (Andreato et al., 2017).

To this regard, both anaerobic alactic and lactic systems are determinant as they define the intensity and capacity of continuous application of effective and powerful strikes (attacks) throughout the match with a high glycolytic contribution (Andreato et al., 2016; Franchini, Cormack, et al., 2018; Gastin, 2001; Vasconcelos et al., 2020). A high aerobic level it is considered beneficial for enhanced recovery, between each high-intensity bout and in between matches (Tomlin & Wenger, 2001).

To cope with this high-intensity intermittent effort pattern and the specificity of physiological demands, it is suggested that BJJ athletes should perform high-intensity interval training (HIIT) sessions varying in terms of exercise modality (general— e.g., running, rowing, and cycling—and sport-specific), time (effort and pause intensities, durations, and ratios) and structure (series and repetitions) (Buchheit & Laursen, 2013; Franchini, Cormack, et al., 2018; Vasconcelos et al., 2020).

During the match Brazilian Jiu Jitsu athletes must sustain the ability to resist against an opposition for a long period of time, by maintaining a strong grip on different body parts and/or *gi* or kimono. It is why there is a large consensus that not only grip strength must be developed as well as grip endurance (Andreato et al., 2017). Given that BJJ fighters report higher perceived exertions in the forearm (Andreato et al., 2015) training this body region is important and must be included in BJJ strength and condition programmes. The same results appear to occur in Judo athletes, when asked during and after the competition forearms and fingers were body regions with higher RPE (Kons et al., 2018). In this study it was possible to observe a decrement in grip isometric strength absolute values after each match along the competition (Kons et al., 2018). Bonitch-Góngora and colleagues

(2012) analysed the effect of blood lactate concentrations on the handgrip strength during judo bouts and they found an inverse correlation between the two, higher lactate concentrations resulted in lower handgrip values, highlighting the importance of this parameter in the competition. Moreover, grip strength and grip endurance levels can distinguish experienced and novice athletes as well as athletes with different competitive levels and weight categories (Andreato et al., 2017; Franchini, Schwartz, et al., 2018).

3. Methods

3.1. Participants

Eighteen male well trained BJJ athletes (mean \pm SD; age 32 ± 7.3 years; body mass 73.75 ± 11.1 kg;) of national and international level participated in this study.

Participants voluntarily assigned to this study, prior to the familiarization signed an informed consent, according to the Conselho de Ética para a Investigação da Faculdade de Motricidade Humana (CEIFMH – nr. 42/2021) and filled a health questionnaire (Physical Activity Readiness Questionnaire, PAR-Q & YOU). Participants were excluded if: 1) any cardiorespiratory or cardiovascular known disease was reported, 2) after any affirmative answer from written PAR-Q & YOU, and 3) measured arterial hypertension with a systolic pressure above 140 mm/Hg and a diastolic pressure above 90 mm/Hg.

Participants were asked to avoid vigorous exercise, alcohol drinking and caffeine 24h before every test and to maintain their regular diet throughout the study. All test sessions were scheduled and performed approximately at the same time of day (± 2 h), to avoid the effects of chronobiological variability on physiological response (Pullinger et al., 2020). All participants concluded the experimental sessions within a period of fifteen days.

Table 1 - Descriptive statistics of the participants characteristics, mean \pm SD.

	N	Mean	Std. Deviation
Age (years)	18	32,0	7,32
Body mass (Kg)	18	73,8	11,07
BJJ experience (years)	18	8,9	4,16
Valid N (listwise)	18		

3.2. Experimental design

This was an experimental study with a randomized crossover design. Three sessions were conducted, two moments of evaluation, one using RSN and the other using RSH-VHL and a first session were all participants performed a maximal graded test and familiarized themselves with testing procedures and VHL technique. All tests were performed on an arm crank ergometer (Lode Angio, Groningen, Netherlands). The participants were seated and with their shoulder joint aligned with the pedal crank axel.

Maximal arm crank ergometer graded test and familiarization

Before the start of the experiment, all subjects participated in a first session to complete a progressive maximal test to assess their peak oxygen consumption ($\dot{V}O_{2\text{peak}}$) and to familiarize themselves with the testing procedures as well as the VHL technique, as it was described elsewhere by Woorons et al. (2017).

Prior the exercise arterial hypertension was measured, if any impairment was not observed participants followed with the familiarization protocol beginning with a three-minute warm-up with no load applied (0W). The first stage began with a load of 15W, and every minute there were 15W increments, until volitional exhaustion was attained. Throughout the test the participants were required to maintain a cadence of 70 ± 5 rpm, as it corresponds to the workload most likely to reduce earlier onset fatigue before central limiting factors are reached with a slow cadence and avoid a greater isometric component for torso stabilization and greater respiratory drive resulting in increased oxygen consumption associated with a faster cadence (Price et al., 2007). All participants were given strong verbal encouragement during the test and were asked to keep their back always against the seat, in order to diminish torso compensation movements.

Peak $\dot{V}O_2$ was then determined as the highest thirty-second average value attained before participants reached exhaustion.

After a period of rest, participants were asked to familiarize themselves with ventilatory hypoventilation at low lung volume technique, which consisted in performing long and deep breaths without forcing while pedalling with no additional load (0W), participants did an end expiratory breath hold and kept it till the first sign or urge to breath, till a point where they could control each inhalation and exhalation, for about five minutes. The next five minutes participants were asked to do the same procedure but to explore a little further the limits while keeping the end expiratory breath hold and accelerate the pedalling rhythm during the breath holds. After this exercises all subjects participated in four sprints while using VHL technique, simulating the experimental protocol.

In the same session all participants did one set of three bench press throws with no additional load.

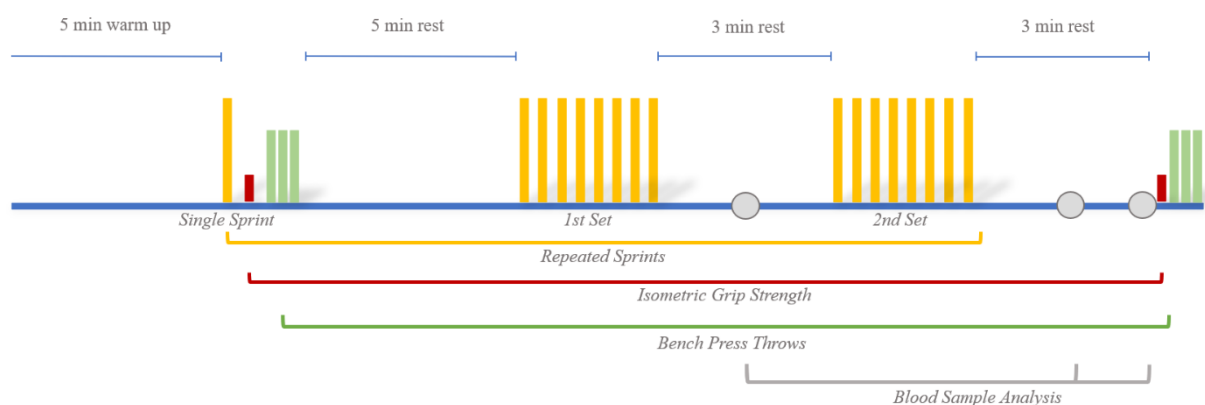
Repeated sprint sessions

After the familiarization phase, all subjects participated in two testing sessions separated by at least 48h within a 15-day period. The testing protocol consisted of two sets of eight “all-out” 6-s sprints on an arm cycle ergometer. The 6-s sprints were separated by 24-s of inactive recovery with departure every 30-s, and 3 min of passive rest, between the two sets (Woorons et al., 2017).

Before starting the first set, subjects performed a 5-min warm-up at low-intensity, pedalling at the arm crank with no additional load, to proceed with the testing protocol, torque was set as the result of body weight times the distance to the crank axel as specified by the manufacturer, the value was given in $N.m^{-1}$. A single 6-s all-out sprint was performed, and peak power output (PPO) was registered and served as a parameter for the first sprint of the first set. Right after the single sprint participants hand grip strength of the dominant hand was measured two times separated with 1 minute rest, participants were asked to stand with the arm extended along the trunk, the athlete was then instructed to generate the greatest possible force during 3–5 sec. Measurements were conducted using a Jamar dynamometer (Jamar, Lafayette, CA, USA) and the higher value obtained was considered. One warm up set of three bench press throws loaded with 25% of the body weight was performed followed by a rest of one minute and thirty seconds, then a second and main set of three bench press throws loaded with 50% of the body weight was performed, peak power was recorded (Chronojump Boscosystems® linear encoder and Chronojump software version 1.9.0.), and the best repetition was kept for analysis.

Exercise began after a 5-min period of rest (Woorons et al., 2017). In the first sprint of the first set (S1), subjects should achieve at least 90% of the peak power output (PPO) reached in the single sprint and if not, they would restart the set again after a 5-min period of rest (Willis, Peyrard, et al., 2019; Woorons et al., 2017). After the repeated sprint protocol participants were asked again to measure isometric grip strength and perform three more bench press throws were performed and peak power recorded in order to assess differences from the unfatigued evaluation.

Figure 3 - Experimental design overview of the repeated sprints training sessions



3.3. Measurements

Performance

For each 6-s sprint, peak power output (PPO), mean power output (MPO) and was measured. Total work (TW) was calculated as a result of the averaged power obtained in each 6-s sprint times the duration of each sprint (6-s). For both sets, fatigue was calculated, using the percentage decrement score (RSA_{decs}) as follows:

$$(100 \times (\text{total sprint MPO} / \text{ideal sprint MPO of the set})) - 100$$

where Total sprint MPO = sum of sprint MPO from all sprints of the set, and Ideal sprint MPO of the set = The number of sprints of the set x highest sprint MPO of the set (Woorons et al., 2017).

Gas exchange and heart rate (HR)

Ventilation and gas exchange parameters were collected by a breath-by-breath gas analyser (MetaMax 3B, Cortex Biophysik, Leipzig, Germany), after calibration according to the manufacturer's instructions. Heart rate was continuously recorded and monitored using an heart rate sensor (Polar® H7, Kempele, Finland). Since gas exchange could not be measured during sprints with breath-holdings in RSH-VHL, data was analysed in the following 20-s of the recovery periods (Woorons et al., 2017). Heart rate was average and analysed over a period of 6-s, the highest value registered during or immediately after each sprint was kept for analysis.

Arterial oxygen saturation (SpO_2)

Arterial oxygen saturation was continuously measured during the first set and the second set using a pulse oximeter placed on the ear lobe (Nonin PureSAT® SpO_2 technology, WristOx2 3150 USB, Plymouth, USA). Data was then averaged and analysed over 6-s, which correspond to the duration of each sprint. lowest value registered during or immediately after each sprint bout was kept for further analysis. It is important to note that, in a cycle ergometer for lower limbs, a SpO_2 drop at the end or just after the 6-s sprints, was reported (Woorons et al., 2017).

Near-infrared spectroscopy (NIRS)

A Niro-200 NIRS instrument (Hamamatsu, Japan) applying three different wavelengths of near-infrared light was used to measure the change in concentration for each sprint for oxyhaemoglobin (O_2Hb), deoxyhaemoglobin (HHb), total haemoglobin (tHb) and tissue saturation index (TOI) (Willis, Peyrard, et al., 2019). Data was averaged and analysed over a 6-s period, the highest value of HHb obtained during the sprint or

just right after it was kept for analysis, the same time period was considered for the other NIRS parameters. The skin was shaved and the NIRS probe was placed on the belly of the triceps brachii long head, given that triceps brachii long head participates in both elbow and shoulder extension, an elastic non-compressive bandage was wrapped around the probe to prevent movement and any source of interference. A pen and photographic record were used to mark the probe placing for later testing.

Rating of perceived exertion (RPE) and blood lactate concentration [La]

In both exercise conditions, just after the end of each set, RPE was obtained using the Modified Borg Scale (range 0–10). Blood lactate concentrations were taken and analysed using a Lactate Pro device (Arkay, Kyoto, Japan). An ear lobe blood sample was taken from the subjects, 90 s after the end of the first (S1) and the second set (S2) as well as 180 s after the end of S2 to obtain [La]. The highest value of the two samples collected at the end of S2 was kept for the analyses (Woorons et al., 2017).

Neuromuscular Fatigue

Isometric hand grip strength was measured right after the warm up before the bench press throws, before and after the repeated sprint protocol, two repetitions were executed with 1 minute rest in between, the highest value was kept for analysis. Measurements were conducted using a Jamar dynamometer (Jamar, Lafayette, CA, USA). A bench press throw was performed before and after the repeated sprint protocol. Two sets of three repetitions were executed right after the single 6-sec all-out sprint after the 5-min warm up, one warmup set with 25% of the bodyweight with 1 minute and 30 seconds rest between sets and a second set with 50% of the bodyweight. Peak power was recorded, and the best repetition was kept for analysis. A Chronojump Bioscosystems® linear encoder and Chronojump software version 1.9.0. was used to measure and record each repetition.

3.4. Statistical Analysis

All the results are expressed as mean \pm standard deviation (SD). Data was first tested for distribution normality using Shapiro-Wilk test and Levene's test for variance homogeneity. If normality was not verified, Wilcoxon signed ranks test was utilized to assess differences. The data was analysed using a mixed "between-within" analysis of variance (ANOVA), for RSH-VHL or RSN. The Bonferroni post hoc procedure was

performed, whenever a significant interaction effect is observed, to localize the difference. T-Test were performed to assess differences if any, time effect, condition effect or interaction was found. *P* values were set at 0.05.

4. Results

4.1. Participants

Table 2 - Participants characteristics and main results, mean \pm SD.

	N	Mean	Std. Deviation
Age (years)	18	32,0	7,32
Body mass (Kg)	18	73,8	11,07
BJJ experience (years)	18	8,9	4,16
$\dot{V}O_{2\text{ peak}}$ (ml.kg ⁻¹ .min ⁻¹)	18	34,1	7,44
HG _{iso} (Kg)	18	51,0	11,63
Valid N (listwise)	18		

Participants were aged 32.0 ± 7.32 years, weighting 73.8 ± 11.07 Kg. BJJ training experience was 8.9 ± 4.16 years of experience. In the maximal arm crank graded test participant's $\dot{V}O_{2\text{ peak}}$ was 34.1 ± 7.44 ml.kg⁻¹.min⁻¹ and maximal isometric grip strength was $51,0 \pm 11,63$ Kg.

4.2. Performance

Performance results in both conditions are presented in table 3.

Table 3 - Performance results in RSN and RSH-VHL, mean \pm SD.

	RSN		RSH-VHL		ANOVA <i>p</i> value		
	S1	S2	S1	S2	T	C	T x C
MPO (W)	$164 \pm 17,4^{*\dagger}$	$159 \pm 15,7^*$	$157 \pm 16,5^\dagger$	$154 \pm 18,1$	<0,01	<0,01	0,51
PPO (W)	$308 \pm 34,3^{*\dagger}$	$297 \pm 31,6$	$287 \pm 46,9^\dagger$	$282 \pm 46,11$	<0,01	0,06	0,02
TW (J)	$983 \pm 103,7^\dagger$	$951 \pm 93,9$	$904 \pm 209,5^\dagger$	$891 \pm 208,4$	<0,01	0,08	0,15
RSA _{decs} (%)	$-6,92 \pm 2,60$	$-7,54 \pm 2,67$	$-8,79 \pm 3,12$	$-7,86 \pm 3,10$	0,79	0,13	0,26

* Significantly different when compared with RSH-VHL $p < 0.05$

† Significantly different when compared with S2 within condition, $p < 0.05$

No significant interaction between sets and conditions was observed. However, analysis of variance showed that MPO in the first set (161 ± 3.91 SE) was greater than MPO in the second set (157 ± 3.87 SE), $p < 0.01$. Paired samples T Test revealed that MPO in the first set in RSN was greater when compared with the MPO of the first set in RSH-VHL, the same was verified in the second set between RSN and RSH-VHL, $p = 0.02$.

PPO in the first set was higher (298 ± 8.39 SE) than in the second set (289 ± 8.31 SE), $p < 0.01$, PPO in RSN was higher when compared with RSH-VHL, in both the first set and in the second set, $p = 0.02$. T Test showed

that RSN PPO in the first set was greater than RSH-VHL PPO in first set but no differences were found in the second set, $p=0.41$.

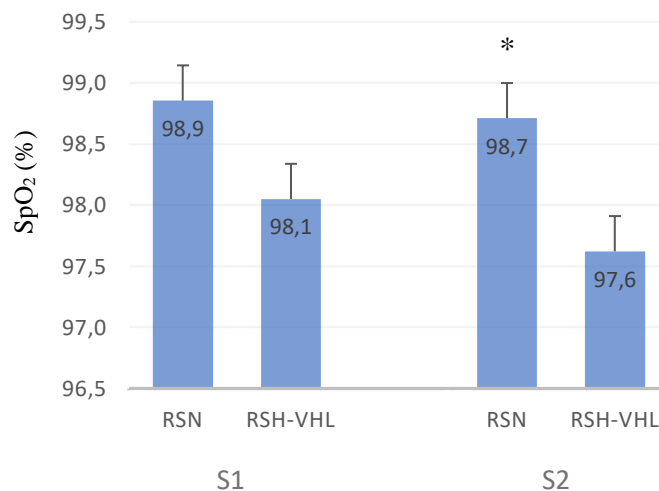
TW was greater in the first set ($944 \pm 33.9 SE$) when compared with the second set ($921 \pm 33.6 SE$), $p<0.01$, but there were no differences between conditions and no interaction between sets and condition.

Percentage decrement score (RSA_{decs}) was not different between sets and conditions, also no interaction effect between the two was observed. There were no significant differences between RSN and RSH-VHL in the first and in the second sets.

4.3. Arterial oxygen saturation and HR

Wilcoxon signed ranks test showed that arterial oxygen saturation was not different between RSN in the first set (98.86 ± 1.50) and RSH-VHL (98.05 ± 1.49), $Z= -1.704$, $p=0.09$, but RSN SpO_2 in the second set (98.71 ± 1.86) was greater when compared with RSH-VHL (97.62 ± 2.60), $Z= -2.249$, $p=0.03$ see figure 4.

Figure 4 - Arterial oxygen saturation results in RSN and RSH-VHL



* Significantly different when compared with 2nd set in RSH-VHL, $p=0.03$

Heart rate results are presented in Table 4. Analysis of variance showed that the first set ($148 \pm 3.13 SE$) was lower than the second ($155 \pm 2.91 SE$) $p<0.01$, RSN ($156 \pm 2.97 SE$) was greater than RSH-VHL ($146 \pm 3.34 SE$) $p<0.01$, but no interaction between the two were observed. A paired samples T Test was performed and revealed that RSN was greater than RSH-VHL in the first set, $p<0.01$ and in the second set, $p<0.01$.

4.4. Gas exchange

Table 4 - Gas exchange and heart rate results in RSN and RSH-VHL, mean \pm SD.

	RSN		RSH-VHL		ANOVA <i>p</i> value		
	S1	S2	S1	S2	T	C	T x C
$\dot{V}O_2$ (ml.kg ⁻¹ .min ⁻¹)	29,0 \pm 3,40 [†]	30,3 \pm 4,17*	29,6 \pm 4,43 [†]	32,6 \pm 5,20	<0,01	0,03	0,02
$\dot{V}E$ (L.min ⁻¹)	68,3 \pm 16,5** [†]	78,9 \pm 20,0	64,0 \pm 13,7 [†]	73,8 \pm 16,4	<0,01	0,03	0,66
RER	1,03 \pm 0,05** [†]	0,98 \pm 0,03	1,00 \pm 0,06 [†]	0,97 \pm 0,03	<0,01	0,02	0,04
HR _{mean} (bpm)	153 \pm 12,4** [†]	160 \pm 12,4*	142 \pm 14,0 [†]	150 \pm 13,1	<0,01	<0,01	0,81

* Significantly different when compared with RSH-VHL, $p < 0.05$

[†] Significantly different when compared with S2 within condition, $p < 0.01$

$\dot{V}O_2$ in the first (29.3 \pm 0.85 SE) set was lower when compared with the second set (31.5 \pm 1.07 SE) $p < 0.01$, and in RSN (29.65 \pm 0.87 SE) $\dot{V}O_2$ was lower than in RSH-VHL (31.12 \pm 1.11 SE), $p = 0.03$, $\dot{V}O_2$ in RSN was lower when compared with RSH-VHL in both the first set (29.0 \pm 0.80 SE vs. 29.6 \pm 1.04 SE) and in the second set (30.3 \pm 0.98 SE vs. 32.6 \pm 1.23 SE), $p = 0.02$. T-Test reveal no differences between conditions in the first set RSN and RSH-VHL, $p = 0.42$ but at the end of the second set, $\dot{V}O_2$ in RSN was lower than in RSH-VHL, $p < 0.01$.

$\dot{V}E$ in the first set (66.14 \pm 3.46 SE) was lower than the second set (76.33 \pm 4.10 SE), $p < 0.01$, and RSN (73.57 \pm 4.24 SE) was higher than RSH-VHL (68.90 \pm 3.50 SE), $p = 0.03$, but no interaction was found between the two, $p = 0.66$. T Test showed that RSN was greater when compared with RSH-VHL in the first set, $p = 0.03$ but there were no significant differences in the second set between RSN and RSH-VHL, $p = 0.07$

RER was greater in the first set (1.02 \pm 0.01 SE) than the second set (0.98 \pm 0.01 SE), $p < 0.01$, RSN (1.01 \pm 0.01 SE) was greater when compared with RSH-VHL (0.99 \pm 0.01 SE), $p = 0.02$, RER in RSN was higher when compared with RSH-VHL in both the first set (1.03 \pm 0.01 SE vs. 1.00 \pm 0.01 SE) and in the second set (0.98 \pm 0.01 SE vs. 0.97 \pm 0.01 SE), $p = 0.04$. Paired samples T Test showed that the first set in RSN, RER was greater than RSH-VHL, $p < 0.01$ but no differences were found in the second set in RSN when compared with RSH-VHL, $p = 0.16$.

4.5. Near-infrared spectroscopy - Muscle Oxygenation

Table 5 - Near infrared spectroscopy results in RSN and RSH-VHL, mean \pm SD.

	RSN		RSH-VHL		ANOVA <i>p</i> value		
	S1	S2	S1	S2	T	C	T x C
O ₂ Hb	-14,9 \pm 8,88	-10,7 \pm 9,68	-16,6 \pm 8,19	-12,4 \pm 8,85	<0,01	0,30	0,92
HHb	27,1 \pm 9,45	28,4 \pm 10,41	27,8 \pm 8,92	28,1 \pm 8,94	0,13	0,89	0,13
TOI	46,7 \pm 11,7	48,5 \pm 11,4	44,4 \pm 10,8	46,4 \pm 10,2	<0,01	0,27	0,87

Near infrared spectroscopy results are presented in Table 5. O₂Hb was greater in the second set (-11.55 \pm 2.05 SE) when compared with the first set (-15.72 \pm 1.84 SE), $p < 0.01$, but no differences were observed between conditions and no interaction between set and condition was found.

There were no differences in HHb between sets, and no differences between conditions, also no interaction was found between both set and condition.

TOI was greater in the second set (47.43 \pm 2.39 SE) when compared with the first set (45.55 \pm 2.44 SE) $p < 0.01$, but no differences were observed between conditions and no interaction was found between set and condition.

4.6. Rating of perceived exertion and [La]

Wilcoxon signed ranks test showed no differences for RPE between RSN in the first set (7.61 \pm 1.29) and RSH-VHL (8.22 \pm 1.59) as $Z = -1.707$ and $p = 0.09$, also no significant differences were observed between RSN in the second set (8.33 \pm 1.53) and RSH-VHL (8.56 \pm 1.15), $Z = -0.665$ and $p = 0.51$.

Table 6 - Lactate results in RSN and RSH-VHL, mean \pm SD.

	RSN		RSH-VHL		ANOVA <i>p</i> value		
	[La] 90-s	[La] max	[La] 90-s	[La] max	T	C	T x C
[La] mmol.L ⁻¹	7,03 \pm 2,13 [†]	10,42 \pm 2,55*	5,64 \pm 1,72 [†]	8,04 \pm 3,00	<0,01	0,01	0,02

* Significantly different when compared with RSH-VHL, $p < 0.05$

† Significantly different between moments within condition, $p < 0.01$

Analysis of variance showed that [La] in the 90-s measurement of the first set (6.34 \pm 0.37 SE) was lower when compared [La] max at the end of the second set (9.32 \pm 0.55 SE), $p < 0.01$. [La] in RSN (8.73 \pm 0.53 SE) was greater when compared with RSH-VHL (6.84 \pm 0.54 SE), $p = 0.01$. [La] in RSN was higher when compared with RSH-VHL in both the first set (7.03 \pm 0.50 SE vs. 5.64 \pm 0.41 SE) and in the second set (10.42 \pm 0.60 SE vs. 8.04 \pm 0.71 SE), $p = 0.04$. Paired samples T Test revealed that [La] measured in the 90 seconds after the first set, in RSN

was greater than RSH-VHL, $p=0.02$. [La] max in RSN was greater when compared with RSH-VHL, $p<0.01$. See all [La] results in Table 6.

4.7. Neuromuscular fatigue

Table 7 - Neuromuscular fatigue results in RSN and RSH-VHL, mean \pm SD

	RSN		RSH-VHL		ANOVA p value		
	Pre	Post	Pre	Post	T	C	TxC
HG _{iso} (Kg)	49,6 \pm 9,85	49,0 \pm 9,39	51,0 \pm 11,63	49,9 \pm 10,56	0,44	0,29	0,72
BP _{PPO} (W)	1504 \pm 477,4 [†]	1428 \pm 445,3	1493 \pm 469,7	1450 \pm 435,0	0,18	0,88	0,41

[†] Significantly different when compared with post value within condition, $p<0.05$

Analysis of variance showed no effects or interaction between time and condition for both bench press throw peak power and isometric grip strength.

5. Discussion

To the best of our knowledge, this was the first study to compare the effects of a repeated sprint protocol with and without the utilization of VHL in an arm cycle ergometer.

This study revealed some significant findings. First, MPO was significantly lower in RSH-VHL when compared with RSN. On the other hand, RSA_{decs} and bench press throw peak power drop after the repeated sprint protocol were not impacted by ventilatory hypoventilation at low lung volume. Second, a significant SpO_2 drop was observed at the end of the second set in RSH-VHL but there were no significant changes in muscle deoxygenation. And third, blood lactate concentration was lower in RSH-VHL when compared with RSN. Finally, RSH-VHL induced an increase in $\dot{V}O_2$ when compared with the same exercise in normal breathing conditions.

In all performance indicators only one interaction effect was identified, in the first set PPO was greater than the second and RSN higher than RSH-VHL. It was also possible to see time effects in MPO and TW, with the first set higher than the second set, and although MPO was greater in RSN when compared with RSH-VHL, there were not significant changes in total work between the two conditions. Decrements in mean power output or performance impairment has already been reported while RSH in normobaric hypoxia when compared with normoxic conditions (Goods et al., 2014; Morrison et al., 2015; Smith & Billaut, 2010), so our findings suggest that special considerations have to be taken into account by coaches and sport physiologists while planning and periodizing whenever they decide to use RSH-VHL over traditional RSE in normoxia. The lower values obtained in MPO in RSH-VHL and the interaction effect observed in PPO, as some authors had anticipated (Woorons et al., 2017), could be due to acute respiratory and metabolic acidosis since it has already been observed in previous studies (Woorons, 2014; Woorons et al., 2007, 2010). This could have led to greater [La] and greater RER, but that was not observed in our study.

Goods et al. (2014), reported a decline in mean power output and SpO_2 during RSH in consequence of gradual increase in simulated altitude. This was also verified by Smith & Billaut (2010). A possible explanation that has been given by several authors, relies with the fact that phosphocreatine resynthesis and H^+ removal are oxygen-dependent processes (Goods et al., 2014; Morrison et al., 2015). It is plausible that this could in part explain our mean power output decrement in RSH-VHL since a SpO_2 drop was verified in our study.

Smith & Billaut (2010), have demonstrated that a decrease in arterial oxygen saturation may also contribute to a central nervous system fatigue during RSH via a cerebral oxygenation decline. In a recent study conducted by Woorons & Dupuy, et al. (2019) and although they found a decline in cerebral oxygenation that

responded faster than the SpO₂ decline curve, it did not negatively affect the RSA decrement score. The authors questioned Smith and Billaut's explanation given that unlike RSH, during the recovery periods following sprints with VHL, SpO₂ rapidly returns to normal levels (or close), which therefore favours the oxidative process of the recovery and consequently maintenance of performance. However, it is important to recognize that Woorons & Dupuy, et al. (2019) did not measure any other performance indicators, such as MPO, PPO or TW so it is difficult to affirmatively reject this hypothesis. Furthermore, it is plausible that even if we did not measure it, that our participants had a similar cerebral oxygenation decline pattern and that it may have had in fact an influence on our PPO and MPO reported values.

Phosphocreatine (PCr) it is known to be of particular importance during RSE, where a high rate of adenosine triphosphate (ATP) utilization and resynthesis is required (Girard et al., 2011). After a maximal 6-second sprint stores can be reduced to about 35-55% of resting levels and to complete recovery of phosphocreatine stores require more than 5 minutes (Girard et al., 2011). Additionally, it has been reported a greater phosphocreatine reduction in fast twitch fibres than in slow-twitch fibres, as fast twitch fibres dominate the power production during a supra maximal exercise such as RSE, selective phosphocreatine deficit of those fibres might be related to a limitation in replicate performance when sprints are repeated (Girard et al., 2011).

Repeated sprint decrement score was not different between sets or protocol condition, this corroborates the findings in cycling (Woorons et al., 2017) and running (Woorons & Dupuy, et al., 2019). This is important because it confirms what has already been observed has one of the main characteristics that favours the use of RSH with VHL over RSH done in normobaric hypoxic environments once the effectiveness of RSH and the positive effect over performance depends on the recruitment of a high number of fast twitch fibres (Woorons & Dupuy, et al., 2019). Moreover, the $\dot{V}O_2$ increase may have favoured the phosphocreatine resynthesis contributing this way to the non-observation of differences in the sprint ability decrement score between conditions.

For the first time in studies that utilize VHL technique we have measured peak power in bench press throw exercise before and after the protocol in order to assess the impact of RSH-VHL protocol when compared with RSN on explosive actions. The bench press throws peak power was not different after the repeated sprint protocol in RSH-VHL but was different within RSN. The results suggest a minimal or none impact whatsoever in bench press throw peak power. The reason may be explained by the action itself and its nature, it is a high intensity explosive movement done in few milliseconds therefore it recruits high threshold fast twitch fibres, that were not fatigued or negatively affected by the repeated sprint protocol in VHL. The results can translate the same reasoning done with RSA_{decs}. As a highly dependent movement in phosphocreatine availability, and since we

observed a higher $\dot{V}O_2$ it is probable that again, favouring a faster phosphocreatine resynthesis during the RSH-VHL protocol and right after it, ATP availability has been provided without decreasing bench press throw peak power after the repeated sprint protocol. The isometric hand grip strength was not changed or impacted by RSH-VHL, the results may be explained by the action of repeated sprinting at the arm cycle ergometer that probably do not exert to much solicitation of the forearm muscles.

Regarding the gas exchange parameters, we found significant changes in $\dot{V}O_2$ between the two protocols and an interaction effect was found between set and condition, $\dot{V}O_2$ was greater in the second set and greater in RSH-VHL, which is in line with previous research (Woorons & Dupuy, et al., 2019; Woorons et al., 2011, 2017). This group of authors attributed this result as a consequence of the O_2 debt incurred during breath holds (Woorons, Dupuy, et al., 2019). And they advanced that the greater oxygen consumption during RSH-VHL could have been sufficient to preserve both muscle and cerebral oxygen delivery and consequently to enable the maintenance of performance compared with RSN (Woorons & Dupuy, et al., 2019). On the other hand, the same authors suggested that the greater fall in arterial and cerebral oxygenation, the higher compensation in $\dot{V}O_2$. Given that HR_{mean} was lower in RSH-VHL and there was no difference in muscle HHb between protocols on one hand, and there was a higher $\dot{V}O_2$ in RSH-VHL when compared with RSN, this increase it is probably due to elevated cardiac output, which would be the consequence of higher stroke volume (Woorons & Dupuy, et al., 2019). An increased stroke volume was previously reported during recovery periods following RSH-VHL (Woorons et al., 2011) as a result of a “pump effect” induced by the large and rapid inhalations that occur as soon as the breath holding ends (Woorons & Dupuy, et al., 2019). The higher $\dot{V}O_2$ in RSH-VHL may also explain the lower blood lactate concentration and lower RER which could favour a better lactate clearance. Woorons’ research group believe that the glycolytic activity is increased with the utilisation of VHL technique since an increase in blood lactate concentration has been observed in the past (Woorons et al., 2011, 2014), however we did not find the same pattern. It is important to acknowledge that rest times and exercise intensities compared were different from the latest repeated sprints protocol, the first protocols researched utilized submaximal intensities and shorter rest times (Kume et al., 2013, 2016; Woorons et al., 2010, 2011) when compared with RSH-VHL protocols (Woorons & Dupuy, et al., 2019; Woorons et al., 2017) which utilized shorter efforts at higher intensities and longer rest periods.

It is known that the mitochondrial metabolism is altered under hypoxic conditions, it seems that cell respond differently over an acute and prolonged exposure to hypoxia. Under prolonged hypoxia (several hours) there is a decrease in cellular respiration (via inhibition of the electron transport chain), which is viewed as a

protective mechanism, and among other effects acid citric cycle is affected which could in turn result in an increased lactate production, on the other hand, during acute hypoxic exposure the electrons transport chain is maintained but is changed resulting in a more efficient transfer of oxygen electrons during hypoxia (Fuhrmann & Brüne, 2017; Lee et al., 2020). Given that during VHL technique hypoxic exposure is interrupted whenever normal breathing is retaken, we can only expect an acute response to hypoxia exposure and therefore a more efficient electron transport chain during oxidative phosphorylation. Considering the importance of phosphocreatine energetic pathway in short duration RSE (Gaitanos et al., 1993), and the higher levels in $\dot{V}O_2$ it is reasonable not to discard the potential effect that this may have positively affected phosphocreatine resynthesis via an augmented oxidative phosphorylation, this may also explain in part the higher levels in oxygen consumption and the lower values on blood lactate concentration and RER.

According with Woorons, Billaut, et al., (2021), HR fall at the end of the repetitions with VHL is a consequence of the large arterial deoxygenation. This has already been observed in previous VHL studies (Woorons, Billaut, et al., 2021; Woorons et al., 2017; Woorons, Lemaitre, et al., 2021) as well as during exercises with breath holding with high lung volume (Ahn et al., 1989 and Lindholm et al., 1999 cited by Woorons, Billaut, et al., 2021) It is considered to be an oxygen-conserving mechanism which results in a decreased cardiac work for reducing the overall oxygen uptake and keep a sufficient oxygen supply to the brain (Woorons, Billaut, et al., 2021). Although we can also consider a primary vagal mechanism behind bradycardic response (Lindholm et al., 1999). Furthermore, after each sprint bout, whenever VHL was applied, participants were encouraged to control and slow down their urge to hyperventilate within the time left to rest with deep and slow breaths, it is therefore highly probable that this may have influenced the autonomous nervous system activity (del Negro et al., 2018) this may have influenced the ventilatory response and may be the explanation why \dot{V}_E was lower in RSH-VHL when compared with RSN.

We were expecting differences between conditions in the NIRS parameters, since just recently Woorons, et al. (2017) reported greater muscle deoxygenation (higher HHb and lower O_2Hb) during the second set of a cycling RSH-VHL compared with the same exercise in normal breathing conditions, but that was not verified in our study, it was only possible to see time effects in O_2Hb and TOI. This means that muscle O_2 extraction and oxygenation was maintained in acute hypoxia despite a reduced O_2 ventilation (Smith & Billaut, 2010). Therefore, this VHL protocol does not seem to compromise O_2 availability within the muscle. No changes in muscle oxygenation have already been seen in previous studies with VHL and with different degrees of hypoxia (Billaut et al., 2013; Smith & Billaut, 2010; Woorons & Dupuy, et al., 2019).

6. Limitations

While it was possible to reveal novel findings and confirm other recent outcomes, we have to acknowledge some limitations in our study. First, the group was not homogenous in terms of competitive level nor training experience more specifically strength training. In search for a more in depth influence of this variable with all the measurements, we did find an interaction effect between mean power obtained and the two conditions (RSN or RSH-VHL) and experience in strength training (experience or no experience) (data not presented), but that only confirms that subjects with greater absolute forces tended to have greater decrements in the power-generating capacity of the recruited fibres due to greater metabolite-induced disturbances (Mendez-Villanueva et al., 2008). No effect or interference was found in other performance variables and fatigue parameters. Moreover, Woorons (2014) adverts that the use of VHL requires high level of training experience, in his words “VHL is a method intended for well-trained athletes”. Second, the NIRS probe placement in triceps brachii long head may have affected the measurement of this variables, we justified this choice by the fact that triceps brachii long head participates in both elbow and shoulder extension, furthermore, we considered the placement in other muscles such as latissimus dorsi or biceps brachii but that could cause practical issues regarding participants discomfort and friction with the probes making it difficult to measure, moreover there were other studies that found no significant alteration in these parameters between RSN and RSH or RSH-VHL, so probably this choice did not interfered with the values reported. Third, the VHL technique requires some practice and training, Woorons (2014) suggest a training programme starting with exercises with no movement and subsequently progressing with the application of VHL at submaximal intensity exercises, culminating with breath holds during repeated sprint exercise, that being said the familiarization phase may have been short and may have influenced the experiment results.

7. Practical applications

VHL technique has been utilized with a wide range of modalities, swimming, cycling, running and here we have exposed the acute effects of RSH-VHL with an arm cycle ergometer. We utilized an effort:pause ratio of 1:4 (6-s:24-s), since MPO and PPO were significantly affected by RSH-VHL protocol and given RSH-VHL seems to provide advantage to the utilization of the phosphocreatine, higher rest times should be considered and/or lower working periods.

The RSA_{decs} and bench press throws peak power were not affected by the repeated sprint protocol indicating that it is possible, apart from concurrent considerations regarding the specificity of the stimuli, to include this protocol in a common training session.

8. Future research

One of our study limitations highlighted the importance of strength training and absolute force in power output development during repeated sprinting exercises. It should be of great interest to know how it really affects and what implications it has in repeated sprint training when combined with ventilatory hypoventilation at low lung volume, physiologically and metabolically. At the other spectrum, how much aerobic conditioning affects RSH-VHL. Also differences between high-level and low-level athletes, in terms of competitive and training experience. Considering gender specificities, it is also important to know how it can differ with female and with male athletes.

It is known that emotional stress response is not only related with performance itself but also to athlete's mental health as well, considering that the VHL technique application involves breathing training exercises and since it is known that respiratory work is related with the autonomic nervous system response, it is highly probable that this could also be one of the advantages of RSH-VHL training in long term.

9. Conclusions

In conclusion this study revealed that although statistically significant the reduction of the arterial oxygen saturation with the utilization of ventilatory hypoventilation with low lung volume may not have been sufficient to induce significant levels of hypoxia. MPO and PPO were significantly affected by RSH-VHL but there was not a significant decrease in total work, suggesting that special considerations have to be taken into account by coaches and sport physiologists whenever they decide to use RSH-VHL over traditional RSN. On the other hand, RSA_{decs} was not affected. For the first time we introduced a fatigue evaluation of explosive movements to analyse the impact of the repeated sprint protocol in both conditions, and it did not reveal any impact or effect. These two parameters combined suggest that VHL technique has not affect the repeated sprint ability and the recruitment of a high number of fast twitch fibres, crucial to the repeated sprint training effectiveness.

Blood lactate concentration and respiratory exchange ratio were lower in RSH-VHL when compared with RSN and $\dot{V}O_2$ was greater. These values combined suggest that ATP-PCr may have been privileged over glycolytic energetic pathway contradicting what has been suggested, via augmented oxidative phosphorylation. This also explain why [La] have been lower in RSH-VHL when compared with RSN.

10. References

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