UNIVERSIDADE ESTADUAL DE CAMPINAS FACULDADE DE EDUCAÇÃO FÍSICA

EDUARDO BODNARIUC FONTES

PERCEPÇÃO DE ESFORÇO EM EXERCÍCIO SOB FADIGA EM NORMÓXIA E HIPÓXIA

Campinas 2011

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Tese de Doutorado apresentada à Pós-Graduação da Faculdade de Educação Física da Universidade Estadual de Campinas para obtenção do título de Doutor em Educação Física na área de concentração Ciência do Desporto.

Orientador: Antonio Carlos de Moraes

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Antônio Carlos de Moraes Orientador

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RESUMO

O presente trabalho buscou um maior entendimento da formação da percepção subjetiva de esforço (PSE) durante esforços exaustivos. Dessa forma, o primeiro estudo verificou as associações da atividade muscular (EMG) com a PSE, bem como a determinação do limiar de esforco percebido (LEP) e de fadiga neuromuscular (LFN). Esse estudo analisou 11 adultos jovens durante testes de carga constante até a exaustão voluntária máxima com monitoramento constante de PSE e EMG. A taxa de aumento dessas variáveis (EMG_{slope} e PSE_{slope}) foram significativamente correlacionados e inversamente associados ao tempo de exaustão. LEP e LFN e não se diferiram significativamente. Assim, indicamos a estreita relação do recrutamento adicional de fibras com o aumento da PSE. O segundo estudo foi realizado durante estágio no exterior (sanduíche) na África do Sul no ano de 2009. Nesse trabalho, foi analisado os efeitos da diminuição de oferta de oxigênio (hipóxia) sobre variáveis centrais e periféricas e suas associações com PSE. Seis ciclistas realizaram testes exaustivos de carga constante em normoxia e hipóxia com contínua aquisição de respostas de PSE. EMG e oxigenação muscular (MOX) e cerebral (COX). Foi demonstrado que na condição hipóxia ocorre um significativo aumento sobre PSE em seus diferentes modos (local, respiração e geral), EMG e COX, mas não em MOX. Os slopes de PSE e valores finais de COX foram relacionados ao desempenho em normóxia, no entanto ainda maiores foram apresentados em hipóxia. Além disso, COX foi ainda significativamente relacionada RPE local em normóxia e novamente, hipóxia exerceu efeitos maiores nessas associações, mas dessa vez para todos os modos de PSE. No terceiro estudo, foram utilizado os mesmos dados do estudo anterior para verificamos os possíveis efeitos de hipóxia ao estimarmos LEP de maneira diferenciada (local, respiração e geral) e LFN pelo mesmo protocolo. Todos os modos de LEP diminuíram significativamente sob hipóxia, com maiores efeitos sobre LEP local. Já LFN não respondeu aos efeitos da condição experimental. Dessa forma, expandiu-se a utilização de LEP para altitudes moderadas e foi apresentado uma nova forma de predizer capacidade aeróbia referente aos membros envolvidos e respiração, além de PET para o corpo como um todo. Associando os achados dos estudos, podemos inferir a estreita relação de respostas periféricas e centrais sobre a formação de PSE, senda essas fortalecidas em condições de diminuídas ofertas de oxigênio. Mais adiante, essas associações justificam a ampliação de utilização prática de PSE, podendo ser para o exercício de alta intensidade ou monitoramento localizado da capacidade aeróbia.

Palavras-Chaves: Oxigenação cerebral; córtex pré-frontal; desempenho humano; hipóxia.

FONTES, Eduardo B. Perception of effort in fatigue in normoxia and hypoxia. 2010. 70f. Tese de Doutorado em Educação Física) - Faculdade de Educação Física. Universidade Estadual de Campinas, Campinas, 2010.

ABSTRACT

The present study aimed to bring better understanding of ratings of perceived exertion (RPE) during exhaustive exercise. Thus, the first study verified the associations of the neuromuscualr responses (EMG) with RPE, as well as the determination of the perceived exertion threshold (PET) and neuromuscular fatigue threshold (NFT). Eleven adults performed exhaustive constant-load tests with RPE and EMG recordings. The rate of increase of these variables (EMG_{slope} e RPE_{slope}) were significantly related and associated to performance. Além disso, PET and NFT did not differed. Therefore, it was shown the close relationship of the additional muscle recruitment and RPE. The second study was completed during the international internship in South Africa in 2009. At this investigation, were demonstrated the effects of decreased fraction of inspired oxygen (hypoxia) on central and peripheral responses, as well their relationship with RPE. Six trained cyclists completed exhaustive constant-load tests under normoxia and hypoxia having continuously monitoring of RPE, EMG and cerebral (COX) and muscle (MOX) oxygenation. It was shown that under hypoxia there is a significant increase for all RPE modes (legs, breathing and overall), EMG and COX, but not MOX. The RPE slopes and end values for COX were related to performance under normoxia, however higher associations were found under hypoxia. In addition, COX was significantly related to RPE for legs under normoxia, but again, hypoxia exert higher effects on this association, but this time to all RPE modes. During the third study, the data from last investigation was used to verify the possible effects of hypoxia when estimating differentiated PET (legs, breathing and overall) and NFT during same protocol. All PET modes decrease significantly under hypoxia, with higher effects of PET legs, however, NFT estimation was not affects by this experimental condition. Thus, PET's used was expanded to moderated altitudes and presented a new method to predict aerobic capacity associated to active limbs and breathing, in addition to whole body PET. Associating the studies' findings it is possible to conclude that there is a strict relationship of peripheral and central responses to RPE construct, being this sthrengthed by decreased oxygen availability. Furthermore, these relationship justifies the practical use RPE, as for prescription of high intensity exercise or localized monitoring of aerobic capacity.

Keywords: brain oxygenation, pre-frontal cortex, human performance, hypoxia.

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LISTA DE SIGLAS E ABREVIATURAS

FEF	Faculdade de Educação Física
PET	Perceived exertion threshold
NFT	Neuromuscular fatigue threshold
PSE	Percepção subjetiva de esforço
PSE _{slope}	Taxa de aumento da percepção subjetiva de esforço
EMG _{slope}	Taxa de aumento de atividade muscular
VO ₂	Consumo de oxigênio
VO _{2max}	Consumo máximo de oxigênio
COX	Oxigenação cerebral
MOX	Oxigenação muscular
VE	Ventilação
RPE	Ratings of perceived exertion
rpm	Rotações por minuto
min	Minutos
RMS	Root-mean-square
%RMS	Porcentagem de root-mean-square inicial
fMRI	Ressonância magnetica functional
NIRS	espectrocopia por infra-vermelho próximo
Pmax	Potência maxima
T88%	Intensidade da carga realizada a 88% de Pmax
T94%	Intensidade de carga realizada a 94% de Pmax
T100%	Intensidade de carga realizada a 100% de Pmax
VL	Músculo Vasto Lateral
RPE-L	Percepção subjetiva de esforço das pernas
RPE-B	Percepção subjetiva de esforço da respiração
RPE-O	Percepção subjetiva do esforço do corpo como um todo
O2Hb	Oxi-hemoglobina
DHb	Deoxi-hemoglobina
THb	Hemoglobina Total
UNICAMP	Universidade Estadual de Campinas

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Introdução

Como percebemos nosso corpo? Calor ou frio? Sede ou fome? Conforto ou desconforto? Alegria ou tristeza? Disposição ou cansaço? Prazer talvez? Sabemos que muitas das nossas sensações são baseadas em respostas fisiológicas de nossos segmentos periféricos e centrais (Kandell, Schwartz e Jessel, 2000), mas, sabemos também, que aspectos cognitivos podem influenciar nossas percepções (Marcora e Staiano, 2010). Perguntas como: o que, quando, onde ou mesmo quem influencia mais nessas percepções talvez nunca sejam respondidas, já que essas respostas são extremamente subjetivas e a individualidade biológica e o aspecto sócio-cultural se inter-relacionam substancialmente na formação de nossas percepções. De qualquer forma, a busca por um maior entendimento entre essas relações fisiológicas e cognitivas é desafiadora.

Os limites físicos do ser humano, bem como, a fadiga, tem sido alvo de inúmeros estudos e muita discussão ao longo dos anos (Enoka, 1992; Noakes, 1997; Bassett e Howley, 1997; Noakes, 1998; Weir et al., 2006; Noakes e Tucker, 2008; Marcora, 2010; Amann e Secher, 2010). Mesmo todo o avanço tecnológico e o desenvolvimento de novos aparelhos de mensuração de respostas biológicas não foram suficientes para apresentar um consenso nesse assunto. Em 1915, Angelo Mosso sugeriu, "fadiga cerebral diminui a força para os músculos". Em seu livro, ("La Fatiga"), ele dedicou capítulos para a fadiga central e mental e sua interação com fadiga periférica e muscular. Em 1924, o ganhador do prêmio Nobel, Dr Hill (e colaboradores), sugeriu que um "governador" no coração ou sistema nervoso controlaria o exercício fatigante (Hill et al., 1924). Curiosamente, no entanto, durante o século 20 a maioria dos pesquisadores de fisiologia do exercício não considerou a importância do cérebro em suas investigações, o que acabou ocasionando várias más-interpretações em relação ao exercício e fadiga (Noakes, 1997).

O esquecido conceito de "governador" durante o exercício foi levantado novamente por Noakes (1998), e tem chamado a atenção da comunidade científica sobre as fortes evidências do papel do cérebro sobre a regulação do exercício (Noakes et al. 2005, Noakes e Marino 2007, Swart et al., 2009, Baron et al., 2009). Estas idéias "contemporâneas" têm questionado os modelos tradicionais de exercício e fadiga que focam suas hipóteses em falha catastrófica de um ou mais sistemas corporais no qual a falta de produção de ATP por vias aeróbia é supostamente associada a baixa oferta de oxigenação aos músculos (Noakes 1997). Essa controvérsia tem gerado inúmeras discussões (Basset and Howley, 1997; Weir et al. 2006; Marcora 2009). Dessa forma, exalta-se a necessidade de investigações que apresentam análises integrativas das respostas fisiológicas e discutam modelos teóricos de fadiga durante o exercício físico.

Alguns estudos têm tentado relacionar a fadiga às respostas cerebrais (Kayser, 2003; Hampson et al., 2001; Noakes et al., 2005, Rasmussen et al., 2009). No entanto, a maioria dessas investigações tem estudado a função cerebral por respostas perceptuais (e.g., percepção subjetiva de esforço) (Eston et al., 2007, Faulkner e Eston, 2007) ou por técnicas que acessam áreas superficiais (i.e., espectrocopia por infra-vermelho - NIRS) (Subudhi et al 2009; Perrey 2009) e ou profundas do cérebro (ressonância magnética funcional - fMRI) (Fontes et al., 2010). Contudo, o papel do cérebro durante o exercício e fadiga continua incerto.

A percepção subjetiva do esforço (PSE) é uma variável psicofisiológica que tem sido amplamente usada para monitorar, prescrever e avaliar a intensidade de esforço em diferentes tipos de exercício e populações (American College of Sports Medicine, 2005; Borg, 1982; Borg, 1998; Coutts et al., 2003; Day et al., 2004; Foster et al, 2001; Groslambert e Mahon, 2006, McGuigan et al., 2008). Estudos apontam a associação de PSE com diferentes respostas fisiológicas (Borg, 1998, Lagally, et al., 2002; Coutts et al., 2009, Fontes et al., 2010). Essa simples variável de alta aplicação prática tem demonstrado também alta relação com o desempenho físico, além da possibilidade de estimar o consumo máximo de oxigênio (VO_{2max}) (Faulkner et al., 2007). Recentemente, extensa discussão tem sido apresentada quanto realmente as respostas subjetivas de percepção do esforço são influenciadas sobre as condições periféricas sinalizadas pelos mecanismos de feedback ao cérebro (Marcora, 2010). Apesar de PSE apresentar um importante link entre os aspectos fisiológicos e cognitivos, sua formação ainda não é consenso na literatura.

Uma interessante estratégia para investigar os mecanismos da fadiga e desempenho humano tem sido a manipulação da oferta de oxigênio durante o exercício físico extenuante (Subudhi et al 2007, Amann et al., 2007; Subudhi et al., 2008). Na realização de atividade física na condição de hipóxia (e.g., altitude), diversas variáveis fisiológicas apresentam respostas diferenciadas à condição normóxia (Subudhi et al., 2007; Noakes, 2009; Wang et al

2010) e importante supressão do desempenho das modalidades esportivas (Kayser, 2003) é observado. Apesar de muitos estudos terem focado seus esforços para descrever os efeitos da hipóxia no exercício físico, poucos apresentaram respostas cognitivas, como a própria PSE. Recentemente, apresentamos um estudo (Fontes et al., 2010) que demonstrou estreita relação entre PSE e atividade muscular (EMG) durante esforços fatigantes, no entanto, essas relações sempre foram realizadas na condição normóxia e, nenhuma variável central (e.g., oxigenação cerebral) foi monitorada.

Dessa forma, o objetivo geral desse trabalho foi avançar no esclarecimento da formação da percepção de esforço em estado de fadiga, seja em normóxia, ou mesmo em hipóxia. Acredita-se que descrevendo melhor os mecanismos centrais e periféricos dessa variável em diferentes condições, sua aplicabilidade prática pode aumentar para um melhor controle e monitoramento da intensidade do exercício.

A presente tese de doutorado é descrita aqui em três capítulos. No primeiro capítulo, é apresentado o artigo "The relationship between rating of perceived exertion and muscle activity during exhaustive constant-load cycling". Nesse trabalho, foi descrito as relações das taxas de aumento de PSE (PSEslope) com a atividade muscular (EMGslope) durante cargas constantes exaustivas, além de estimar e comparar os limiares de esforço percebido (LEP) e fadiga neuromuscular (LFN). Esse trabalho foi recentemente publicado no International Journal of Sports Medicine (Oct;31(10):683-8, 2010). Já no segundo capítulo, é apresentado um estudo realizado durante o estágio de doutoramento na África do Sul sobre a supervisão do Dr Timothy Noakes. Nesse trabalho, buscou-se o entendimento dos efeitos da diminuição da oferta de oxigênio sobre as respostas cerebrais e periféricas e suas associações na formação de PSE e desempenho. E por último, com um objetivo mais voltado a aplicação prática dos achados, foi investigado a determinação de LFN e LEP com respostas dos membros ativos, respiração e geral sobre hipóxia.

CAPÍTULO 1

THE RELATIONSHIP BETWEEN RATING OF PERCEIVED EXERTION AND MUSCLE ACTIVITY DURING EXHAUSTIVE CONSTANT-LOAD CYCLING

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ABSTRACT

The aims of this study were to verify the relationship between rating of perceived exertion (RPE) and electromyography (EMG) increases during exhaustive constant-load cycling bouts and, to compare and to correlate the power outputs corresponding to perceived exertion threshold (PET) and neuromuscular fatigue threshold (NFT). Eleven men completed three to four different exhaustive constant-load cycling bouts on a cycle ergometer, being RPE and EMG measured throughout the bouts. The linear regression of the RPEslope and EMGslope against the power output identified the PET and NFT intensity, respectively. There was a significant relationship between RPEslope and EMGslope (R²=0.69; P<0.01). However, the linearity of RPEslope (R2=0.93\pm0.07) was significantly higher (P<0.001) than EMGslope (R²=0.63\pm0.25). In addition, the RPEslope and EMGslope were related to time to exhaustion (r = -0.59 and r = -0.60; P<0.001). There was no significant difference (P=0.42) between PET (201.5±27.9W) and NFT (210.3±22.6W) and they were significantly correlated (r=0.78; P=0.005). Therefore, the RPE and EMG increases during exhaustive constant-load cycling bouts are related and, PET and NFT intensities are similar and closely associated.

INTRODUCTION

The rating of perceived exertion (RPE) is a psychophysiological variable that has been used to monitor and prescribe the intensity of different exercise modes for healthy individuals (American College of Sports Medicine, 2005; Borg, 1998; Coutts *et al.*, 2003; Day et al., 2004; Foster *et al.*, 2001), children and elders (Groslambert e Mahon, 2006), obese (McGuigan *et al.*, 2008), and diseased people (Eston e Collony, 1996). The RPE is related to physiological variables such as heart rate, oxygen consumption and blood lactate concentration (Borg, 1998, Coutts *et a.*, *l* 2003, Lagally *et al.*, 2002). In addition, RPE responses during resistance training (Lagally *et al.*, 2002, 2004) and endurance performance (Garcin *et al.*, 1998) are associated with the degree of skeletal muscle recruitment measured by electromyography activity (EMG).

Studies have shown that cycling at fixed or self-selected intensity (moderate exercise intensity) and fixed exercise duration may not change EMG activity (Duc et al., 2005; Lucia *et al.*, 2000). However, exercise performed at high intensity and till exhaustion constantly requires additional and progressive muscle fiber recruitment to compensate the force loss associated with muscle fatigue, thereby increasing EMG activity (DeVries et al., 1982; Moritani et al., 1993). The recruitment of additional muscle fibers may also be associated with an increased RPE (Enoka, 1992), and it has been suggested that the response of the latter is regulated by the central nervous system (Marcora et al., 2008; St Clair Gibson et al., 2006; Ulmer et al., 1996). In addition, several studies have shown a linear increase in the EMG (DeVries et al., 1982, Hill, 1999) and RPE (Nakamura et al., 2005a; 2005b; 2008; 2009) during exhaustive constant-load exercise, which allows the estimation of their increasing rates (EMGslope and RPEslope, respectively), however, the relationship between these two variables has not been investigated. The confirmation of the significant relationship between EMGslope and RPEslope during exhaustive constant-load exercise will reinforce RPE as a simple tool to predict and monitor exercise intensity, and, it may also be considered an indirect measure of muscle activity during such exercise bouts.

Using similar protocol and linear regression, the RPEslope and the EMGslope have been used to determine the perceived exertion threshold (PET) and the neuromuscular fatigue threshold (NFT), respectively (Moritani *et al.*, 1993, Nakamura *et al.*, 2005). The PET

seems a reliable aerobic index (Nakamura et al., 2009) due to its similarity and high correlation with critical power (r=0.87-0.98) and maximal oxygen consumption steady state intensity (r=0.92) (Nakamura et al., 2005a; 2005b; 2008; 2009), as well as the bias and limits of agreement between PET and critical power are acceptably low (Nakamura et al., 2009). On the other hand, NFT has been related to anaerobic threshold (r=0.82; 0.92) (Matsumoto et al., 1991; Moritani et al., 1993) and rowing performance (r=0.96) (Maestu et al., 2006), although its validity as an aerobic index has been questioned (Maestu et al., 2006; Pavlat et al., 1995). Theoretically, both PET and NFT represent similar phenomena, that is, a steady state of perceptual and neuromuscular responses (i.e., zero slope), respectively, throughout long lasting exercise (Moritani et al., 1993; Nakamura et al., 2008; 2009). Nevertheless, the PET and NFT have not been directly compared. If these methods predict similar exercise intensities, PET should clearly have an advantage over NFT, since simpler procedures are required to evaluate exercise performance. Furthermore, PET can be used to evaluate training effects and control the exercise intensity during fitness programs. For instance, the reduction of the RPEslope of an individual at a given power output would indicate improvement in aerobic performance. Finally, although the EMG may also be useful for such practical application, RPE is more attractive since technical devices are not required to perform the tests.

Therefore, the aims of this study were: (1) to verify the relationship between RPE and EMG increases during exhaustive constant-load cycling bouts and (2) to compare and correlate the power outputs corresponding to PET and NFT.

METHODS

Participants

Eleven physically active (~2-3 recreational exercise sessions per week) nonsmoking healthy men (23.4 \pm 5.2 years; 73.6 \pm 5.1 kg; 177.8 \pm 7.0 cm) participated in this study. They were instructed to refrain from vigorous activities and ingestion of beverages containing alcohol or caffeine in the 24-h prior to each test. This study was approved by the local Institutional Research Ethics Committee and has been performed in accordance with the ethical standards of this journal (Harriss e Atkinson, 2009). In addition, the participants were informed about the procedures and risks before giving written consent to participate in the study.

Experimental design

This study was conducted within a 3-week period, in which participants reported to the laboratory on 3-4 occasions with a minimum of 48-h between sessions. They were all fully familiarized with the tests and experimental procedures, since they had already participated in previous studies using similar protocols. The tests consisted of 3-4 different high-intensity constant-load bouts performed randomly until exhaustion on a cycle ergometer. The number of tests completed varied according to participants' availability to return to the laboratory (4 tests: n=8; 3 tests: n=3). Such a variation in the number of sessions was allowed due to 3-4 predictive tests have not influenced on the PET and NFT determination (DeVries *et al.*, 1982; Hill, 1999; Nakamura *et al.*, 2005). During all predictive tests, RPE, EMG and power output were recorded to determine PET and NFT.

Predictive tests

The tests were performed on an electronically braked cycle ergometer (Quinton Corival 400, Lode Medical Technology, Groningen, Holland). Seat and handlebar height were individually recorded during the first test and reproduced in the subsequent ones. Prior to each test, participants warmed up by cycling at 50 W for 3 minutes followed by 2 minutes of passive recovery. During the tests participants were instructed to maintain a cadence of 60 rpm. Test interruption (i.e., exhaustion) occurred when participants were unable to sustain a cadence greater than 55 rpm for a period of 5 s, despite strong verbal encouragement. To accomplish the exhaustion time target (i.e., ~1 to 15 min), we have empirically determined in our laboratory that relative power outputs should lie within 2.5 and 4.5 W per kilogram of participant's body mass. This procedure was adapted from Hill *et al.* (1999). In spite of this, three participants had one of the predictive tests longer than 15 min. All tests were completed approximately at the same time of the day. No feedback concerning the power output or elapsed time was provided to the participants during the tests.

Determination of perceived exertion threshold (PET)

The Borg 6-20 scale (Borg, 1998) was displayed in front of the participants during all tests. Instructions about reporting their RPE were given before each predictive test, with anchoring as follows: "number 7 represents unloaded cycling while number 19 indicates an

exertion similar to exhaustive cycling". Participants were asked to accurately report their whole body feelings (i.e., overall RPE) (Faulkner e Eston, 2007) every 30 second period. The RPE scores generated from these tests were plotted against time (independent variable), and linear regression indicated the slope coefficient (RPEslope) (Figure 1a). The PET intensity was defined as the x-intersection of the regression line for the power output from the predictive tests and its respective RPEslope (Nakamura *et al.*, 2005a; 2005b; 2008; 2009) (Figure 1b).

Determination of neuromuscular fatigue threshold (NFT)

Active bipolar (20 mm center-to-center) surface electrodes (TSD 150TM, Biopac Systems®, CA, USA - common mode rejection ratio: 95 dB) were used to measure vastus lateralis muscle activity from the participant's dominant leg. The electrodes were positioned between the motor point and the proximal tendon (Hermens *et al.*, 2000). Inter-electrode impedance was minimized by careful skin shaving and alcohol cleaning. The reference electrode was placed over the anterior iliac crest. Ink landmarks were made around the electrodes so that they could be placed in a constant position for all tests. The EMG signal was amplified (MP150 Electromyogram Amplifier, Biopac Systems Inc, Santa Barbara, Ca. USA) and applied a frequency band filter ranging from 20 to 500Hz. The EMG signal was digitized with a sampling frequency of 2000 Hz and processed by calculating the root-mean-square (RMS) every 5 s (AcqKnowledge 3.8.1 TM software, Biopac Systems®, CA, USA). The EMG was normalized to the initial 5 s of each trial. The NFT was estimated by determining the increase rate for the total exercise period (EMGslope) for each predictive test (Moritani *et al.*, 1993) (Figure 1c). The slopes were plotted against their respective power outputs. The NFT was obtained as the x-intercept of the linear regression (DeVries *et al.*, 1982) (Figure 1d).



Figure 1. Estimation procedure of perceived exertion threshold (PET) and neuromuscular fatigue threshold (NFT) of a participant. 1a: Ratings of perceived exertion (RPE) plotted as a function of time for 4 workloads. 1b: Rate of increase of RPE (RPE_{slope}) from 1a plotted for each of the 4 intensities and the projected slope zero (PET). 1c: Percentage of root-mean-square (%RMS) plotted against time. 1d: Rate of increase of %RMS (%RMS . s⁻¹) from 1c plotted for each of the four intensities and the projected slope zero (NFT).

Statistics

Descriptive statistics are presented as mean \pm standard deviation, unless otherwise stated. Least square linear regression was used to fit the data in order to estimate EMGslope and RPEslope. Data normality and homogeneity of variance were confirmed. The coefficients of determination between EMGslope and RPEslope were used to identify their relationship. The t-test for paired samples was used to compare PET and NFT power outputs and the coefficients of determination (R²) associated with data fitting. Pearson product-moment was used to verify the correlations between PET and NFT, as well as among EMGslope and RPEslope with performance (i.e., time to exhaustion). The bias and limits of agreement (LoA) of PET and NFT were calculated using Bland-Altman analysis [3]. The significance level was set at P < 0.05. Data were analyzed using a statistical software (SPSS for Windows, version 17). The power outputs performed by the participants in the predictive tests ranged from 190 to 340 W and time to exhaustion from 105 to 1500 s. Table 1 depicts the results of RPEslope, EMGslope, PET, NFT and their respective coefficient of determination. The linearity of RPEslope was significantly higher than for EMGslope (P < 0.001) and a significant relationship between these variables was found ($R^2 = 0.69$; P < 0.01), as shown in Figure 2.

Table 1. Rate of increase of ratings of perceived exertion (RPE_{slope}) and of electromyography activity (EMG_{slope}), perceived exertion threshold (PET), neuromuscular fatigue threshold (NFT), coefficients of determination (R^2) and correlation of RPE_{slope} and EMG_{slope} with time to exhaustion.

	RPE _{slope} (units.s ⁻¹)	R ²	EMG _{slope} (%RMS.5s ⁻¹)	R ²	PET (W)	R ²	NFT (W)	R ²
mean	1.50	0.93	1.56	0.63*	201.5	0.91	210.3	0.94
SD	0.97	0.07	1.24	0.25	27.9	0.11	22.6	0.04
relationship with performance	-0.59 [†]		-0.60 [†]					

* significantly different from R^2 of RPE_{slope} (P < 0.001).

† significant correlation (P < 0.01).



Figure 2. Relationship between the increasing rate of ratings of perceived exertion (RPE_{slope}) and electromyography (EMG_{slope}) for all work-loads (P < 0.01).

Table 2 shows the individual power outputs for NFT, PET, and their respective standard errors (SE) and R². No significant difference was observed between PET and NFT (201.5 ± 27.9 W and 210.3 ± 22.6 W, respectively; P = 0.42), and their SE and R2. In addition, a significant relationship between PET and NFT was found (r = 0.78; P < 0.01). Furthermore, Figure 3 depicts the results of the Bland-Altman 95% LoA analysis between PET and NFT. The bias was -8.9 ± 17.5 W and the LoA ranged from 25.4 W (+1.96 SD) to -43.1 W (-1.96 SD), evidencing a good agreement.

		NFT		PET			
n	P (W)	SE (W)	R ²	W	SE	\mathbf{R}^2	
1	193.6	9.2	0.95	212.1	14.0	0.95	
2	222.1	12.1	0.98	209.0	25.9	0.92	
3	196.3	13.7	0.98	175.1	53.0	0.79	
4	192.5	17.5	0.90	200.7	9.2	0.97	
5	213.7	24.9	0.93	225.4	16.6	0.96	
6	227.1	7.8	0.96	211.7	16.1	0.90	
7	244.2	12.1	0.92	237.8	10.8	0.94	
8	168.7	19.8	0.89	151.8	4.6	1.00	
9	232.6	25.2	0.47	203.0	33.8	0.61	
10	197.8	7.8	0.98	161.2	14.0	0.97	
11	227.0	16.5	0.89	229.6	7.8	0.97	
mean	210.5	15.2	0.89	210.6	18.7	0.91	
SD	22.4	6.2	0.15	27.8	14.1	0.11	

Table 2. Individual power output (P) of neuromuscular fatigue threshold (NFT) and perceived exertion threshold (PET), and their respective standard error (SE) and coefficient of determination (\mathbb{R}^2).



Figure 3. Bias ± LoA 95% accessed by Bland-Altman analysis from perceived exertion threshold and neuromuscular fatigue threshold.

DISCUSSION

The main findings of the present study confirmed our hypothesis that there is a significant relationship between the EMGslope and RPEslope during exhaustive constant-load cycling bouts, and that both variables predicted a similar intensity for fatigueless prolonged exercise (NFT and PET, respectively). Although other physiological variables have shown a relationship with RPE (Borg, 1998; Eston *et al.*, 2007; Lagally *et al.*, 2004), this is the first study to show a significant relationship between EMGslope and RPEslope during exhaustive constant-load cycling and similarity of the NFT with PET power outputs.

The force loss caused by the fatigued muscle fibers during the constant-load high intensity exercise requires additional motor units' recruitment (DeVries *et al.*, 1982; Moritani *et al.*, 1993), which seems associated with increased RPE (Enoka, 1992). Our data corroborate these studies, evidenced by the significant relationship between EMGslope and RPEslope ($R^2 = 0.69$) during the exhaustive constant-load cycling. While previous studies have shown a corresponding increase in the RPE and EMG during resistance exercise (Housh *et al.*, 2005; Lagally *et al.*, 2004) and cycling using different loads (Macdonald, Farina e Marcora, 2008), a correlation was only shown during leg extension exercise (r = 0.55) (Duncan, Al-Nakeeb e Scurr, 2006) in which the fatiguing effect was not analyzed. The present study expands those finding, demonstrating that the rates of EMG and RPE increase are also correlated during constant-load cycling to exhaustion.

There are a number of different explanations for the link between RPE and EMG responses during fatiguing exercise. Marcora and colleagues (Marcora, Bosio e Morree, 2008; Marcora, 2009) suggested that the RPE during exercise is generated by corollary discharges of the motor commands to the active skeletal muscles. According to this explanation efferent copies of these motor commands are sent to the sensory areas of the brain from where the RPE is processed. In this model, RPE is therefore part of the feedforward control during exercise. On the other hand, Noakes (Noakes, 2009) proposed that the increased skeletal muscle recruitment is the main cause of several physiological responses. The consequences of the increased metabolic demand are sensed by the brain via feedback, thereby raising the RPE (Noakes e Tucker, 2008). The discussion about whether the feedback influences on RPE response

is beyond the scope of this study. However, our results are compatible with both theoretical models since EMGslope and RPEslope were strongly related.

The RPE and EMG responses have been shown to increase linearly until exhaustion during high intensity constant-load exercise (Moritani, Takaishi e Matsumoto, 1993; Noakes, 2008). But, to identify the best fatigueless intensity predictor, the present study compared the linearity of both variables in function of time and showed that the RPEslope ($R^2 = 0.93$) was significantly higher than the EMGslope ($R^2 = 0.63$), however, no differences between their SE and R^2 were found when used to estimate PET and NFT. Despite the differences in data fitting to linear function, these variables present a significant relationship ($R^2 = 0.69$) and similar correlations to time to exhaustion (r = -0.59 and -0.60, respectively).

However, one may question the reliability of the vastus lateralis muscle as a representative muscle of the quadriceps, since different EMG responses have been reported to similar cycling exercising protocol (Dorel *et al.*, 2009; Graef *et al.*, 2009). Although a number of studies reported an increased EMG activity of the vastus lateralis during constant-load high intensity cycling exercise and used to determine NFT (Camic *et al.*, 2010; Graef *et al.*, 2009; Housh *et al.*, 1995; Nakamura *et al.*, 2005; Pavlat *et al.*, 1993) it was recently reported that vastus lateralis and vastus medialis EMG activity did not change during similar exercise protocol (Dorel *et al.*, 2009). These contrasting results may be explained based on different pedal cadence between studies (Hug e Dorel, 2006), since competitive cyclists have a certain pedaling skill regarding the positive recruitment of knee flexors (i.e., biceps femoris muscle) up to the higher cadences, which would contribute to a decrease in peak pedal force and alleviate muscle activity for the knee extensors (e.g., vastus lateralis and vastus medialis muscle) (Takaishi *et al.*, 1998). Then, a lower pedal cadence in the present study (~60 rpm) compared with the other one (95±8 rpm) (Dorel *et al.*, 2009) may explain the increased EMG activity of the vastus lateralis muscle in our study.

In the present study, the NFT and PET power outputs were similar, significantly correlated (r = 0.78; P < 0.01) and showed acceptable agreement (bias = -8.9 ± 17.5 W). These findings can be explained by the close association between the EMGslope and the RPEslope and similar procedures for their estimations. While NFT has been criticized as it overestimates well established aerobic capacity indices (DeVries *et al.*, 1982; Maestu *et al.*, 2006; Pavlat *et al.*, 1995), PET is equivalent to critical power and highly correlated with ventilatory

threshold (Nakamura *et al.*, 2009). However, the estimation of the fatigueless intensity by NFT using EMG requires expensive equipment and personal expertise. In contrast, PET can provide similar power output estimation by using subjective responses from a single scale, even though familiarization to the procedure might be required.

One limitation when estimating the fatigueless intensity by PET, as well as NFT, is the number of tests (~3-4) exhaustive tests needed throughout different days. However, identifying changes in a single RPEslope on a specific power output might help to monitor training effects. For instance, after a training period, the RPEslope during an exhaustive constant-load cycling exercise can decrease when compared with a previous one. In such a case, the individual may perceive less effort when exercising at the same intensity, indicating fitness improvement. However, this suggestion has to be experimentally investigated for further conclusions. Moreover, Nakamura and colleagues (Nakamura *et al.*, 2008) proposed a method to estimate PET by non-exhaustive bouts, at which the RPEslope ranging from 14-17 of the Borg's scale provided similar intensities when compare to PET with all RPE responses. These aspects may improve the practical application of RPEslope and PET.

In conclusion, our results presented a significant relationship between the increase in RPE and EMG activity during exhaustive constant-load cycling bouts. In addition, PET and NFT showed similar power outputs, standard errors and coefficients of determination. Then, both techniques seem to be a good predictor of the fatigueless intensity for prolonged exercise, although the PET can be more attractive since technical devices (i.e., EMG) are not required to perform the tests. The data provided new information regarding the physiological meaning of RPE and PET, and how these parameters are related to neuromuscular aspects during exercise. Furthermore, RPE may be useful to control intensity during exhaustive cycling exercise, providing an indirect measure of the muscle activity.

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CAPÍTULO 2

RELATIONSHIP OF PERCEIVED EXERTION WITH CEREBRAL OXYGENATION DURING EXHAUSTIVE EXERCISE IN HYPOXIA

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ABSTRACT

This study verified the effects of hypoxia on rating of perceived exertion (RPE), peripheral and central physiological parameters, as well as on their relationship during exhaustive constant-load exercise. Six trained male cyclists $(30.2\pm5.7 \text{ years}; 78.9\pm11.3 \text{ kg}; 181\pm10 \text{ cm}; 61.6\pm5.7 \text{ years}; 78.9\pm11.3 \text{ kg}; 181\pm10 \text{ cm}; 61.6\pm5.7 \text{ years}; 78.9\pm11.3 \text{ kg}; 181\pm10 \text{ cm}; 61.6\pm5.7 \text{ years}; 78.9\pm11.3 \text{ kg}; 181\pm10 \text{ cm}; 61.6\pm5.7 \text{ years}; 78.9\pm11.3 \text{ kg}; 181\pm10 \text{ cm}; 61.6\pm5.7 \text{ years}; 78.9\pm11.3 \text{ kg}; 181\pm10 \text{ cm}; 61.6\pm5.7 \text{ years}; 78.9\pm11.3 \text{ kg}; 181\pm10 \text{ cm}; 61.6\pm5.7 \text{ years}; 78.9\pm11.3 \text{ kg}; 78.9\pm11.3 \text{ k$ mlO2/kg/min) performed an incremental test for peak power output (PPO) and three constant workload cycling tests at 88% (T88%), 94% (T94%) and 100% (T100%) of PPO under normoxia (FIO₂ = 20.93% FIO₂) and hypoxia (FIO₂ = $15\%O_2$). Slopes of RPE for legs (RPE-L), breathing (RPE-B) and overall (RPE-O), muscle activity (EMG_{slopes}) as well cerebral (COX) and muscular (MOX) oxygenations parameters were assessed. All RPE_{slope} modes and EMG_{slopes} were significantly higher (P < 0.05) in hypoxia compared to normoxia at T88% and T94%, but not (P > 0.05) at T100%. Deoxyhemoglobin (DHb) for COX increased (P<0.05) within hypoxia, but no changes for MOX were observed. Performance in normoxia was only related to RPE_{slopes} (r=-0.57~-0.59; P<0.05), but under hypoxia relationship increased (r=-0.75~0.79; P<0.01), EMG_{slopes} (r=-0.57;P<0.05) and final responses of DHb for COX (r=0.85; P<0.05). In addition, DHb for COX were related to RPE-L_{slope} under normoxia (r=0.83; P<0.05) and strongly associated to the slopes of RPE-L, RPE-B and RPE-O (r=0.77, 0.97, 0.79, respectively; P < 0.01). Therefore, we may conclude that the relationships between perceived exertion with muscle activity, and oxygenation parameters for muscle and pre-frontal cortex, in particular under hypoxic condition, might suggest the interaction of feedback and feedforward on an integrated central regulated exercise control.

INTRODUCTION

Rating of perceived exertion (RPE) is a psychophysiological construct thought to represent the conscious/verbal manifestation of effort that results from interpretations of several physiological and psychological signals integrated in the central nervous system (St Clair Gibson e Noakes, 2004; Noakes, 2008; Tucker, 2009). RPE has been widely used in clinical and sports settings to monitor and prescribe exercise intensity as well for predicting maximal oxygen uptake and exercise performance (American College of Sports Medicine, 2005; Borg, 1998; Eston *et al.*, 2008). Nevertheless, the roles of putative mediators of perceived exertion in different exercise paradigms remain under debate, since peripheral and central mechanisms seems to be involved (Marcora *et al.*, 2011; Noakes, 2011; Marcora *et al.* 2009; Hampson *et al.* 2001).

It been consistently shown that RPE rises linearly as a function of time during constant load exercise (Noakes, 2008; Nakamura *et al.*, 2005; Eston *et al.*, 2007), being the rate at which it increases (i.e., RPE slope) a strong predictor of time to exhaustion when exercise intensity cannot be reduced (Crewe *et al.*, 2008; Pires *et al.*, 2011; Fontes *et al.*, 2010). Thus, increasing attention has been devoted to its related physiological responses during constant load exercise which significant relationships have been reported with cardiorespiratory responses (Pires *et al.*, 2011) and muscle activity (EMG) (Fontes *et al.*, 2010). Together, these results suggest that peripheral physiological responses contribute, at least in part, to the rate of increase in perception of effort and tolerance to exercise, however, the influence of central parameters such as brain oxygenation needs to be further investigated (Subudhi *et al.*, 2009).

Faster RPE increases in experimental conditions are known to shorten exercise duration including previous fatiguing activity (Eston *et al.*, 2007), hot environment (Crewe *et al.*, 2008) and reduced muscle glycogen stores (Noakes *et al.*, 2004; Lima-Silva *et al.*, 2010) are considered to support the idea that the central command output (i.e., RPE) is set as a function of remaining exercise time, being part of a feedfoward/feedback mechanism (Noakes *et al.*, 2004, Crewe *et al.*, 2008). Decreased exercise performance has also been widely shown under conditions of reduced oxygen availability, (i.e., hypoxia) (Clark *et al.*, 2007; Amann *et al.*, 2007; Romer *et al.*, 2007). Despite the ongoing debate about the multiple peripheral and central mechanisms explaining these responses (Amann e Kayser, 2009), reductions in pre-frontal cortex oxygenation (COX) during strenuous exercise have been associated with impairments in central

motor drive (Subudhi *et al.*, 2009; Goodal *et al.*, 2010; Rasmussen *et al.*, 2010) and decision to stop exercise (Subudhi *et al.*, 2009). Furthermore, there are evidences that skeletal muscle activity (Kayser *et al.*, 1994), muscle oxygenation (MOX) (Romer *et al.*, 2007) and perceived exertion, (Maresh *et al.*, 1993) responses to exhaustive exercise are also affected by the fraction of inspired oxygen (FIO₂), but to our knowledge none study concurrently addressed the effects of hypoxia on the RPE, EMG, MOX and COX responses during exhaustive constant-load dynamic exercise, as well as verify their relationship.

In addition to overall RPE, perceived exertion can be differentiated in central (cardio-respiratory, breathing or chest) and peripheral (local, legs or muscular) components, which in turn are considered to be coupled to specific physiological mediators related to the chest and the working limbs, respectively (Robertson e Noble 1997; Maresh *et al.*, 1993; Pandolf, 1982; Robertson, 1982). Although differentiating RPE slopes could provide insightful information on the perceptual signal dominance during whole body dynamic exercise, there is scarce data on this topic (Lima-Silva *et al.*, 2010). In hypoxia, an increased perception of effort for breathing has been commonly reported during intense exercise (Cibella *et al.*, 1999; Lane *et al.*, 1990), however, the influence of this condition on lower limbs perceived exertion has been under investigated (Maresh *et al.*, 1993). Furthermore, the estimation of the rate of increase of overall and differentiated RPE under normoxia and hypoxia, as well as their association with peripheral (e.g., MOX, EMG) and central (i.e., COX) responses might help to better understand the mechanisms involved on RPE estimations and reinforce this simple method as an interesting tool to prescribe specific training intensities and improve its practical application under different inspired fraction of oxygen, as in altitudes up to 2500 m (FIO₂ 15%).

Therefore, the purposes of this study were to verify the effects of hypoxia on the slopes of RPE, EMG, and MOX and COX, as well as on their relationships with performance during exhaustive constant-load exercise. We hypothesize that reduced time to exhaustion in constant load exercise under hypoxia would be accompanied by greater slopes of RPE, EMG, MOX and COX, which in turn will be strong related to each other in both normoxic and hypoxic conditions.

METHODS

Subjects

Six trained male cyclists $(30.2 \pm 5.7 \text{ years}; 78.9 \pm 11.3 \text{ kg}; 181 \pm 10 \text{ cm}; 61.6 \pm 5.7 \text{ mlO}_2/\text{kg/min}; 8.3 \pm 2.2 \text{ training years}; 6.0 \pm 3.1 \text{ training hours per week}, 316 \pm 209 \text{ km per week})$ signed an informed consent and volunteered to take part in this study after being properly instructed about the purposes, procedures, risks and benefits of the investigation. Athletes were instructed to avoid exhaustive exercise 24h hour preceding all trials and to not ingest caffeinated substances neither alcoholic drinks during the experimental period. All subjects were well familiarized to the experimental procedures since they had already participated in previous studies using similar methods. In addition, they were used to complete exhaustive high intensity efforts during their day-by-day cycling training routine. The study protocol was approved by the local ethics committee and completed according to the declaration of Helsinki.

Experimental design

This study was completed within an 8-week period in which subjects reported to the laboratory on seven occasions separated by at least 48 h. An incremental test was performed followed by three different intensities constant-load exhaustive cycling bouts completed under normoxia and hypoxia. Each athlete was tested at approximately the same time of the day with ambient temperature ranging from 21 to 23°C and 60% of humidity. During the tests, differentiated ratings of perceived exertion, as well as, cardiorespiratory parameters and muscle activity were assessed. Time to exhaustion (i.e., performance) and plasma lactate were collected at the end of each test. For each inspired oxygen condition, the peripheral and central physiological variables were calculated for comparisons with overall and differentiated RPE.

Incremental and constant-load tests

For the incremental test, after 5min free chosen cadence and intensity warm up, subjects started the test at an initial workload of 100W and 20W increments per minute were applied as a ramp matter until exhaustion. Peak power output (PPO) and peak of oxygen uptake (VO_2) were accessed.

During the constant-load tests, three bouts were performed under nomoxic $(FIO_2 = 20.93 \%)$ and three under hypoxic $(FIO_2 = 15 \%)$ conditions. Exercise intensities were set at 88% (T88), 94% (T94) and 100% (T100) of PPO and were completed on a counterbalanced order. After a five minutes sitting rest baseline period with the respective FIO₂, a five minute warm up at 100W was completed. Then, an additional 2 minutes rest to adjust the respective workload and subjects started cycling until exhaustion. Time to exhaustion was taken to the nearest second. In all tests, athletes used their own bicycles attached to a cycling simulator (Computrainer Pro, Racer Mate, Seattle, USA) and were instructed to maintain the cadence at ~85 rpm. Exhaustion was defined as the incapacity to keep the cadence for more than five seconds. Strong verbal encouragement was provided by the same researcher during all tests. Subjects were blinded to the exercise intensity performed and completed time duration during the trials. One subject was injured and did not complete the T88% tests under both conditions.

Inspired O₂ concentration

An altitude simulator (AltiTrainer200, SMTEC, Switzerland) was used to control the normobaric hypoxic condition (FIO₂ 15%). This equipment dilutes the ambient air with nitrogen via a mixing chamber, with the dilution being constantly controlled by a PO₂ probe. Four meters long tube was connected to a valve (2700 Two-way non-rebreathing valves T-ShapeTM, Kansas, USA) and attached to the VO₂ mask. In order to blind the subjects to the O2 supply condition they used the same facemask during the normoxic and hypoxic trials without information whether it was connected or not to the altitude simulator. In addition, a small fan was attached to the latter to simulate the subject's ventilation and produce the similar characteristic noise during the normoxic trials.

Ratings of perceived exertion

During the constant load trials subjects reported their ratings of perceived exertion for the legs (RPE-L), breathing (RPE-B) as well as the overall RPE (RPE-O) using Borg's 6-20 scales (Borg, 1982). Overall and differentiated scores were taken every 30 seconds on a randomized order at each reporting time point. Before each test standard instructions were provided on the RPE anchoring as follows: "number 7 represents unloaded cycling while number

19 indicates an exertion similar to exhaustive cycling, please, when asked, separately report your exertion for legs, breathing and overall to your whole body exertion". RPE scores were then plotted against time in each exhaustive trial and the resulting linear regression angular coefficients were taken as the slopes for overall (RPE- O_{slope}), legs (RPE- L_{slope}) and breathing (RPE- B_{slope}) perceived exertions. In addition, slope was also established for differentiated RPE responses plotted in function of percentage of completed time to exhaustion under both conditions.

Muscle activity

For EMG recordings, two electrodes (Blue Sensor, Medicotest, Denmark) were carefully taped 20 mm apart and parallel to the muscle fibers over the Vastus Lateralis (VL) muscle on the subjects' left limb. After shaving and cleaning the regions with ethanol before each test, the VL electrode was placed at two thirds of the distance between the anterior iliac crest and lateral border of patella (Hermens *et al.*, 2000). Electrodes were then connected to a telemetric EMG system (TeleMyo 2400T G2, Noraxon, Arizona, USA) and data was collected at 2000 Hz and band pass filtered at 20-500 Hz using the software MR-XP 1.07 Master Edition (Noraxon, Arizona, USA). Five seconds EMG data was treated as root-mean-square (RMS) windows and normalize to the initial five seconds period of the respective test. Individual EMG slopes for the VL (EMG_{slope}) were obtained for each trial by the regression angular coefficients found between EMG and time (DeVries *et al.*, 1982; Maestu *et al.*, 2006).

Cerebral and muscular oxygenation

A DYNOT near-infrared spectroscopy equipment (NIRx Medical Technologies, New York, USA) was used to assess cerebral (COX) and muscular (MOX) oxygenation parameters. This system measures brain activity in response to stimuli by detecting real-time oxygenated (O₂Hb), de-oxygenated (DHb) and total (THb) blood flow patterns at a depth of 4cm from the head surface, by harmless infrared spectrum light transmitted through laser diodes placed on the scalp. Each diode, simultaneously, emits and detects the dual wavelength light for oxy- and de-oxy hemoglobin (760nm and 830nm, respectively). The procedure was initiated by placing a lightweight adjustable helmet on the head of participants. Four diodes were inserted in the helmet and were held in place by ferrules fixed at a distance of 40mm². Diodes

were fitted around FP1 (EEG map) in order to assess the left pre-frontal cortex. Two other diodes were placed on the right VL muscle, separated by 20mm and positioned over the muscle belly and parallel to muscle fibers. An additional diode was connected to a phantom measuring head as a "one optical reference" channel. The diodes cables were properly attached to support holders in order to facilitate the subjects' movements during the tests. Data sets were acquired at a sampling rate of 1.8Hz and at a dynamic signal range of ~180 dB (1:109) on a host computer using Pacific Scientific OC950 motor control software (RAD motion), and software for instrument control and data acquisition (DYNOT, version 3.0). Near Infrared Analysis, Visualization and Imaging (NAVI version 2.1; NIRx Medical Technologies, 2008), Statistical Parametric Mapping (SPM version 5, 2005) and MRIcro (version 1.4) softwares were used to investigate NIRS data. Data was filtered using a band pass filter with a high pass cut-off frequency of 0.3 Hz and normalized to "co-located/SD channel" using NAVI. The last 10 seconds of each test was used for comparisons between conditions and correlations with performance. In addition, with the linearity of the responses showed by COX and MOX, their slopes were estimated by the regression angular coefficients found between these parameters and time. The initial 30s from each MOX data was removed from the analysis to eliminate the initial disturbances from hemodynamic responses. COX and MOX data from T88% and T94% was removed from the analysis due to technical and operational problems.

VO₂ and lactate

Ventilatory parameters were continuously monitored and averaged over eight respiratory cycles through all tests using a breath-by-breath gas analyzer (K4b2, COSMED, Rome, Italy). The system was calibrated according to the manufacturer's instructions before each test with a $15\% O_2$ and $6\% CO_2$ reference gas mixture and a 3 L syringe. Peak oxygen uptake during all trials and conditions was considered the highest value attained.

At the third and fifth minutes after exercise cessation 100 µl blood samples were collected from subject's ear lobes for posterior determination of plasma lactate concentration (YSI 2300 STAT PlusTM, Yellow Springs, USA), being the highest value [lac]_p considered for analysis.
Statistics

Statistical analysis was carried out using a statistical software package (SPSS for Windows, SPSS Inc., Chicago, USA), being the results presented as median \pm semiinterquartile range (SIR). Since Shapiro Wilk's test confirmed the non-normality of data set, nonparametric tests were employed for inferential analysis. Comparisons between selected variables accessed in different exercise intensities under normoxia and hypoxia were performed by the Friedman's test followed by Wilcoxon's test when pertinent. Relationships among physiological, perceptual and performance parameters were verified by the Spearman correlation coefficient. Since pooled data confirmed data normality, Pearson was applied. In all cases statistical significance was set at P < 0.05.

RESULTS

Variables accessed at exhaustion at different exercise intensities under normoxic and hypoxic conditions are presented in table 3. Times to exhaustion (T_{EX}) and VO₂ attained under hypoxia were significantly lower (P < 0.05) than those under normoxia for all exercise intensities, whereas muscle activity was significantly higher under hypoxia in T88%. No significant differences were found for plasma lactate, overall and differentiated RPE between conditions. Moreover, T_{EX} at T88% under normoxia and hypoxia were different from T94% and T100% (P < 0.05), and T94% also differed from T100% (P < 0.05), however, no differences were found for VO₂ among intensities under both conditions. In addition, VO₂ peak attained during the incremental test was significantly different from T88% and T94% both under hypoxia (P < 0.05).

Table 3. Times to exhaustion, physiological and perceptual parameters at the end of exhaustive tests at 88 (T88%), 94 (T94%) and 100% (T100%) of peak power output under hypoxia and normoxia.

	T88%		Т9	4%	T100%		
	normoxia	hypoxia	normoxia	hypoxia	normoxia	hypoxia	
$T_{EX}(s)$	378.0 ± 29.5	$256.0 \pm 28.0*$	$240.5 \pm 31.0^{\#}$	$180.0 \pm 22.1^{*^{\#}}$	$176.5 \pm 17.3^{\#\ddagger}$	$136.5 \pm 7.9^{*^{\#\ddagger}}$	
O ₂ (mlO ₂ /kg/min)	57.7 ± 8.5	$53.4 \pm 6.0*$	56.2 ± 8.9	$51.4 \pm 3.4*$	61.6 ± 7.6	$52.8 \pm 7.0^{*}$	
[lac]p (mmol/l)	12.8 ± 1.1	15.2 ± 1.2	13.4 ± 1.0	14.5 ± 0.4	13.9 ± 0.4	15.1 ± 0.8	
EMG(%)	23.2 ± 16.9	$41.5 \pm 10.0*$	23.9 ± 11.3	41.3 ± 29.6	$58.6 \pm 18.3^{\#}$	53.0 ± 23.6	
RPE-L	20 ± 0.5	20 ± 0.5	19 ± 0.4	19 ± 1.1	19 ± 0.8	19 ± 0.4	
RPE-B	19 ±0.5	20 ±0.5	19 ±0.4	19 ±0.4	19 ±0.4	19 ±0.4	
RPE-O	20 ± 0.5	19 ±0.5	19 ±0.4	19 ±1.5	19.5 ±0.4	19 ±0.5	
COX DHb (Δ%)					44.0 ± 10.9	70.7 ± 26.9	
MOX DHb ($\Delta\%$)					96.3 ± 47.4	100.5 ± 48.4	
COX THb (Δ%)					-12.5 ± 75.2	-4.0 ± 44.5	
MOX THb ($\Delta\%$)					-47.3 ± 30.4	-1.4 ± 19.1	
/O ₂ (mlO ₂ /kg/min) [lac]p (mmol/l) EMG (%) RPE-L RPE-B RPE-O COX DHb (Δ%) MOX DHb (Δ%) COX THb (Δ%)	57.7 ± 8.5 12.8 ± 1.1 23.2 ± 16.9 20 ± 0.5 19 ± 0.5 20 ± 0.5	$53.4 \pm 6.0*$ 15.2 ± 1.2 $41.5 \pm 10.0*$ 20 ± 0.5 20 ± 0.5 19 ± 0.5	56.2 ± 8.9 13.4 ± 1.0 23.9 ± 11.3 19 ± 0.4 19 ± 0.4 19 ± 0.4 19 ± 0.4	$51.4 \pm 3.4*$ 14.5 ± 0.4 41.3 ± 29.6 19 ± 1.1 19 ± 0.4 19 ± 1.5	61.6 ± 7.6 13.9 ± 0.4 $58.6 \pm 18.3^{\#}$ 19 ± 0.8 19 ± 0.4 19.5 ± 0.4 44.0 ± 10.9 96.3 ± 47.4 -12.5 ± 75.2 -47.3 ± 30.4	52.8 ± 7.0^{3} 15.1 ± 0.8 53.0 ± 23.6 19 ± 0.4 19 ± 0.4 19 ± 0.5 70.7 ± 26.9 $100.5 \pm 48.$ -4.0 ± 44.5 -1.4 ± 19.1	

 T_{EX} = time to exhaustion; VO_2 = oxygen consumption; [lac]p = plasma lactate; EMG_{VL} = vastus lateralis activity; ratings of perceived exertion for legs (RPE-L), breathing (RPE-B) and overall (RPE-O); n = 5 for T88% and n = 6 for T94% and T100%. *P < 0.05 from the same intensity in normoxia; [#]P < 0.05 from T88%; ^{*}P < 0.05 from T94%.

Overall and differentiated RPE_{slope} rose with increasing exercise intensities in both normoxia and hypoxia, being higher values observed in the latter condition (Figure 4A-C). All RPE_{slope} modes were significantly higher (P < 0.05) in hypoxia compared to normoxia at T88% and T94%, but not (P > 0.05) at T100%. Comparisons between intensities revealed RPE slopes at T100% to be significantly higher (P < 0.05) than those at T88% in both conditions, while overall RPE slope at T100% under hypoxia was also greater (P < 0.05) than that at T94%. Within the same intensity and condition overall RPE slope was significantly higher (P < 0.05) than RPE slope for the legs under hypoxia at T100%.



Figure 4. RPE slopes for A: legs (RPE-L_{slope}), B: breathing (RPE-B_{slope}) and C: overall (RPE-O_{slope}) during exhaustive constant load exercise bouts at 88% (T88%), 94% (T94%) and 100% (T100%) of peak power output performed under normoxia and hypoxia. n = 5 for T88% and n = 6 for T94% and T100%. *P < 0.05 from the same intensity under normoxia; [#] P < 0.05 from T88% under same condition; ^{*} P < 0.05 from RPE-L at same intensity and condition.

EMG slope values at different exercise intensities under normoxia and hypoxia are presented in figure 5. EMG slopes increased as a function of exercise intensity with greater values observed under hypoxia. Significant differences between conditions (P < 0.05) were found at T88% and T94%, whereas comparisons among intensities indicated EMG slopes at T100% to be significantly greater (P < 0.05) than those at T88% for both conditions.



Figure 5. EMG slopes of vastus lateralis during constant bouts performed until exhaustion at 88% (T88%), 94% (T94%) and 100% (T100%) under normoxia and hypoxia. n = 5 for T88% and n = 6 for T94% and T100%. *P < 0.05 from the same intensity in normoxia; [#]P < 0.05 from T88% in the same condition;

Cerebral and muscular oxygenation parameters are shown on figure 6. When compared to baseline, O_2Hb for COX and MOX showed a decrease pattern at the end of the exercise and DHb an increase. For THb, COX presented reduced median values and for MOX, an augmented approach. When comparing the slopes of cerebral and muscular oxygenation parameters between conditions accessed at T100%, only DHb was higher in hypoxia then in normoxia (P < 0.05), as shown by figure 6 (A-B).



Figure 6. Cerebral (COX) (A) and muscular (MOX) (B) oxygenation parameters slopes during constant bouts performed until exhaustion at 100% under normoxia and hypoxia. O2Hb = oxyhemoglobin; DHb = deoxyhemoglobin; THb = total hemoglobin); n = 6 for T100%; *P < 0.05 from the same parameter in normoxia.

In addition, for pooled data RPE slopes were also significantly related to T_{EX} under normoxia (r = -0.57~-0.59; P < 0.05) and hypoxia (r = -0.75~0.79; P < 0.01) (Table 4). On the other hand, EMG slope was significantly related to T_{EX} under hypoxia (r = -0.57; P < 0.05) but not normoxia (r = -0.46; P > 0.05). When verifying the relationship of time to exhaustion of T100% with the final values of cerebral and muscular oxygenation parameters, no significant associated variables were found for MOX in both conditions and COX in normoxia, however, for

COX in hypoxia, moderated correlations were found for O_2Hb (r = -0.52) and THb (r = -0.72) and significantly values for DHb (r = 0.85; P < 0.05).

Table 4. Relationship of time to exhaustion with overall and differentiated perceived exertion slopes and final cerebral and muscle oxygenation parameters during exhaustive constant-load under normoxia and hypoxia.

							COX			MOX	
	RPE-L _{slope}	RPE-B _{slope}	RPE-O _{slope}	EMG _{slope}	_	O ₂ Hb	DHb	THb	 O ₂ Hb	DHb	THb
T _{EX}											
normoxia	-0.59*	-0.57*	-0.58*	-0.46		-0.02	-0.75	-0.19	0	-0.03	-0.04
T_{EX}											
hypoxia	-0.79**	-0.75**	-0.77**	-0.57*		-0.52	-0.85*	-0.72	 -0.03	-0.13	-0.18

 T_{EX} = time to exhaustion; RPE-L_{slope}, RPE-B_{slope}, RPE-O_{slope} = slope of ratings of perceived exertion for legs, breathing and overall, respectively; O₂Hb = oxyhemoglobin; DHb = deoxyhemoglobin; THb = total hemoglobin; Cerebral parameters compare with time to exhaustion only at T100%; *P < 0.05; **P < 0.01

Figure 7 shows the relationships between different RPE slopes and EMG slope under normoxia and hypoxia. EMG slope was significantly related to all RPE slopes in both conditions, with higher coefficients of determination observed under hypoxia (r = 0.81 to 0.90; P < 0.01) compared to normoxia (r = 0.72 to 0.82; P < 0.05).



Figure 7. Relationship of the slope of ratings of perceived exertion for legs (RPE- L_{slope}), breathing (RPE- B_{slope}) and overall (RPE- O_{slope}) with the slope of muscle activity of Vastus Lateralis (EMGVL_{slope}) during constant and high intensity cycling under normoxia and hypoxia.

When RPE slopes were expressed relatively to the percentage of completed time to exhaustion, no significant differences were found for the comparison between conditions (normoxia *vs* hypoxia) and neither among intensities (T88%, T94% and T100%). RPE-L_{slope} under normoxia varied 0.07 to 0.08 unit.%T_{EX} and in hypoxia 0.08 to 0.11 unit.%T_{EX}. For RPE-

 B_{slope} in normoxia were from 0.06 to 0.09 unit.% T_{EX} and 0.08 to 0.13 unit.% T_{EX} in hypoxia. And, for RPE-O_{slope} under normoxia varied from 0.06 to 0.09 unit.% T_{EX} and in hypoxia from 0.08 to 0.11 unit.% T_{EX} .

Table 5 depicts the relationship of RPE and EMG slopes with COX and MOX parameters under normoxia and hypoxia. No significant correlations were found for none of the parameters for MOX in any condition, however, DHb for COX in normoxia had significant correlations with RPE-L (r = 0.83; P < 0.05) and under hypoxia, this parameters was strongly associated to the slopes of RPE-L, RPE-B and RPE-O (r=0.77, 0.97, 0.79, respectively; P < 0.01).

Table 5. Relationship of RPE slopes for legs, breathing and overall (RPE-L, RPE-B and RPE-O, respectively) with oxygenation parameters of cerebral (COX) and muscular (MOX) under normoxia and hypoxia.

	normoxia					 hypoxia							
	MOX			COX		MOX				COX			
	O ₂ Hb	DHb	THb	O ₂ Hb	DHb	THb	 O ₂ Hb	DHb	THb		O ₂ Hb	DHb	THb
RPE-L	0.00	0.21	-0.12	-0.64	0.83*	-0.60	 0.46	0.65	0.64		-0.64	0.77*	-0.03
RPE-B	0.21	0.15	-0.20	-0.75	0.77	-0.71	0.06	0.46	0.20		-0.12	0.97**	0.23
RPE-O	0.21	0.15	-0.21	-0.75	0.77	-0.71	0.20	0.25	-0.06		-0.55	0.79*	-0.29
EMG	0.29	0.29	-0.60	-0.43	0.25	-0.37	-0.48	-0.25	-0.07		-0.60	0.77*	-0.37

 $(O_2Hb=Oxyhemoglobin; DHb = Deoxyhemoglobin; Total hemoglobin = THb). * = P<0.05. **=P<0.01.$

DISCUSSION

The main findings of this study partially confirmed our hypothesis by showing that the slopes of overall and differentiated RPE, EMG and COX (DHb) were augmented in hypoxia during exhaustive constant-load cycling but not for MOX. Although it has been recently reported that RPE and EMG slopes to be related in healthy subjects (Fontes *et al.*, 2010), the present study extends these findings for well trained cyclists and differentiated RPE slope, which have also showed correlations with EMG and COX slopes, as well as with time to exhaustion under normoxic (P<0.05) and hypoxia conditions (P<0.01). In addition, significant relationship was also found for COX with performance in normoxia, but stronger association was observed

under hypoxia. Thus, our data shows that with decrease oxygen availability and additional muscle recruitment might exert important influence on perceived of exertion.

Given its great practical applicability, much attention has been devoted to RPE construct in exercise science (Borg, 1998). Considered to represent the integration a myriad of physiological and psychological responses during exercise (St Clair Gibson *et al.*, 2006), thus setting the tolerance limit (Noakes, 2008), perceived exertion has been widely shown to be a strong predictor of performance. In this context, intensive research has been conducted in an attempt to elucidate the suggested mechanisms underlying RPE during exhaustive exercise. Hence, different physiological variables are emerging as potential mediators of such parameter, including heart rate, oxygen uptake, respiratory rate, ventilation, blood lactate, ph, mechanical strain and core temperature (St Clair Gibson *et al.*, 2006). Our results showed that during the constant-load bouts performed until exhaustion as used, peak oxygen uptake, plasma lactate and RPE values attained at the end of the bouts attest for the exhaustive nature of such efforts, and in line with prior studies fatigue-related studies (Lima-Silva *et al.*, 2010; Crewe *et al.*, 2008), exercise tolerance expressed by the time to exhaustion was reduced in our volunteers under hypoxia. Irrespective of the intensity, performance was decreased by ~ 30 %.

Amann and colleagues (2006) showed that EMG activity increased linearly in function of negative slope of the oxygen blood saturation. Therefore, the decreased delivery of oxygen to the muscle might had induced higher rates of muscle recruitment (i.e., EMG_{slope}), as also shown by our data. And consequently, higher rates of central command output (i.e., RPE_{slope}) would be necessary to activate fresh motor units in order to compensate the force loss from the fatigued ones (Moritani *et al.*, 1993., DeVries *et al.*, 1982) and maintain the required exhaustive constant-load exercise. Together, the association found between RPE_{slope} and EMG_{slope} parameters under hypoxia in the present study could be explained by these mechanisms. Although related increases in motor unit recruitment and perception of effort could be interpreted as a causative, this assertion should be viewed with caution given that others physiological and psychological factors are shown to affect RPE and exercise performance (Marcora *et al.*, 2009; Crewe *et al.*, 2008; Lima-Silva *et al.*, 2010), in particular in hypoxic conditions (Kayser *et al.*, 1996).

Decreased prefrontal oxygenation has been shown to impair exercise performance (Goodal et al., 2010; Subudhi et al., 2009). Since regional blood flow and

oxygenation are directly correlated with neuronal activity, the reduction in prefrontal oxygenation may have, at least in part, influenced the ability to maintain motor output. This hypothesis is supported by the fact that prefrontal oxygenation is directly correlated with maximal hand grip strength and is a strong of predictor of motor performance (Rasmussen et al., 2007). In addition, Subudhi and colleagues (2009) indicated that these responses are also accompanied and related by oxygenation reduction in motor and sensory cortex during exhaustive exercise. These authors suggested that these areas may act together in order to orchestrate the decision to terminate exercise. Marcora and colleagues (Marcora, Bosio and Morree, 2008; Marcora, 2009) proposes that RPE is exclusively estimated by corollary discharges from motor to sensory cerebral cortex, independent of any feedback. Therefore, the decreased prefrontal cortex oxygenation with close relationship with RPE under both fraction of inspired oxygen could partially be explained with Marcora's theory. In addition, Williamson and colleagues (2001) showed that during constantload exercise, manipulations of RPE by hypnoses caused changes in respiratory parameters which suggest the role of feedforward on cardiovascular control without any peripheral influence. However, this same author has recently suggested the need of a more integrative system to explain RPE formation. On this direction, a different theoretical form has been proposed by Noakes (1998; 2005; 2011). Despite the large discussion about its validity among all kinds of exercise, the central governor model has gained attention during the last few years (Noakes, 2011). This theory suggests that several physiological responses from periphery and central nervous system are integrated in the brain and RPE would act as a conscious awareness of exercise regulation. Thus, exercise under deleterious conditions imposed by environment, such as the decrease availability of inspiring oxygen used on the present study, RPE would be constructed by peripheral (e.g., EMG, MOX) and central (e.g., COX) information and increase in higher and still linear rates until maximum tolerance of exercise is reached with consequent prevention of catastrophic failure of vital organs (Tucker, 2009; Noakes, 2011). In addition, carotid and brain O₂ sensing has been show to constantly interact with several brain areas and control ventilator responses (Powel et al., 2009). Thus, the decreased COX in hypoxia associated with higher rates of muscle recruitment (r = 0.77; P < 0.05), RPE (r = 0.77 ~ 0.97; P < 0.01) and performance (r = -0.85; P < 0.05) shown by our data suggests the close interaction of peripheral and central physiological parameters to construct RPE, especially with decreased FIO₂. Despite the strong arguments proposed by Marcora (2009) which the feedforward (e.g., teleoanticipation) seems to play an important role on RPE formation, our data showed that the feedback influence must not be dismissed, as also commented by us elsewhere (Smirmaul *et al.*, 2010). Furthermore, Craig (2009) showed in detailed the feedback pathway for pain in the brain, in particular the insular cortex, and indicated the role of perceptual feelings on homeostatic control by the autonomic nervous system. This same author suggests that from a system perspective, the central integration of afferent sensory feedback is designed to support different types of physical activity with different purposes, which each required unique responses to meet the body's physiological needs (Craig, 2003). Therefore, the integrated central regulation model proposed by Noakes's theory, might better explain our results.

Smirmaul (2010) argued that most of the disagreement about the role of feedback on RPE among the researchers is due to terminological problems and instructions when applying RPE scale. Marcora (2011) suggests that when reporting RPE, subjects must answer the following question "*how hard exercise is?*" and any hedonistic response (e.g., pain; discomfort) should excluded to the response. On the opposite way, the proponent of the RPE scale Borg (1982) suggested that whole body feelings must be integrated to indicate the perceived exertion, and differentiated RPE from their specific motor or cardiovascular exertion (Pandolf *et al.*, 1982). We believe that asking the subjects to report their specific information from periphery (e.g., RPE-L, RPE-B) and integration for whole body (RPE-O) as done in the present study would cause no differences, since the exercise protocol (i.e., exhaustive constant-load) would induced parallel changes in both, discomfort and effort during exhaustive exercise. However, further studies with different exercise protocols aiming to separate these feelings should be investigated to better explain this phenomenon.

Prior studies showed higher rates of RPE increase under adverse conditions such as previous fatiguing activity (Eston *et al.*, 2007), hot environment (Crewe *et al.*, 2008, Nybo and Nielsen, 2001) and reduced muscle glycogen stores (Noakes *et al.*, 2004; Lima-Silva *et al.*, 2010), but when this RPE slopes were plotted in function of percentage of time to exhaustion, no differences were found between the conditions. When verifying the effects of hypoxia on these analyzes, our results indicated the same phenomenon with no differences between conditions and intensities. Noakes and colleagues (Tucker and Noakes 2009; Noakes and Marino, 2007; 2011) suggest that the teleoanticipation (e.g., central command) phenomenon would explained this responses. This property was previously reported by Ulmer (1996), that associated

the concept of teleoanticipation to the existence of an extracellular controller of the sustainable metabolic rate during exercise which the organism would act to protect different physiological systems (as the cardiovascular and nervous systems) from possible damage, by decreasing of efferent motor impulses, in anticipation of any risks to cellular homeostasis. This extracellular control would be modulated by subconscious portions of the brain with the feeling of fatigue, one of the main source of information to set effort tolerance. This suggestion should explain the significant correlations of RPE slopes with performance under normoxia (P<0.05) and especially under hypoxia (P<0.01). Decreased O₂ saturation is probably one variable that represents the most obvious threat to homeostasis, whereas during normoxia, COX will only be one of many variables influencing RPE. Since EMG is an indirect measure of brain muscle recruitment and heavily influenced by COX (Subudhi et al., 2009), one would expect that the correlation between EMG and RPE would similarly be better during hypoxia than during normoxia. Furthermore, prefrontal function has been suggested to mainly be responsible for important cognitive functions by integrating internal goals, orchestrate behavior, task related memory, sensory information and motivation to plan motor movements (Rammani and Owen, 2007; Miller and Cohen, 2001). This information could sustain prefrontal cortex as an important part of the central controller. On the other hand, the lower relationship of MOX to performance and RPE_{slope} at this type of strenuous exercise under both conditions, could indicated the important contribution of ATP production from anaerobic sources, as shown by the elevated lactate levels found at the end of the bouts.

Regarding the differentiated RPE responses, Maresh and colleagues (1993) showed no significant differences between overall and legs RPE when low-altitude-landers were exposed to 2200m altitude and performed several exercise intensities. However, our results showed that decreased oxygen availability induced higher rates of perceived exertion for whole body than for legs during the higher performed intensity (100% of PPO). Despite the lack of studies investigating differentiated RPE under hypoxia influence, we hypothesized that oxygen sensing directly in the brain could had imposed an alert feeling about the higher rates of cerebral deoxygenation found in the actual study and anticipate the exercise limits by integrating the whole body perception of effort, and not only for legs. Thus, overall RPE again would act in order to safely set whole body tolerance limit.

Regarding the practical application of the present study findings, with the association of differentiated and overall RPE with peripheral and central physiological responses,

as well as with performance under normoxic and hypoxia, indicate the validity of this parameter to monitor and prescribe exercise intensity in well trained cyclists with different fraction of inspired oxygen, such as in moderated altitude. Furthermore, overall and differentiated RPE might also help by a simple manner verify physiological adaptations provided by exercise training programs.

We acknowledge some limitations of our study. NIRS penetrations depth is limited to superficial areas of the cerebral cortex (3-4 cm deep). This is sufficient to acquire oxygenation parameters only from cortical gray matter, however profound areas related to motor, respiratory, cardiovascular and temperature regulation, such as the basal ganglia, brain stem and cerebellum remain inaccessible by NIRS. Future studies with new brain imaging technologies (e.g. fMRI) are necessary to analyze whole brain activity during exercise and relate to perceived exertion responses.

CONCLUSIONS

In conclusion, our results extend those from previous studies in showing that RPE, COX and EMG slopes are enhanced under exhaustive cycling exercise under hypoxia. Also, that the relationships between perceived exertion with muscle activity, and oxygenation parameters for muscle and pre-frontal cortex, in particular under hypoxic condition, might suggest the interaction of feedback and feedforward on an integrated central regulated exercise control. Together, these data support RPE as a useful marker of the muscle activity both in normoxia and moderate hypoxia, thus being an interesting alternative for monitoring and prescribing exercising intensity.

CAPÍTULO 3

DIFERENTIATED PERCEIVED EXERTION THRESHOLD AND NEUROMUSCULAR FATIGUE THRESHOLD ESTIMATED IN NORMOXIA AND MODERATED HYPOXIA

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ABSTRACT

This study verified the effects of hypoxia on the determination of the differentiated rating of perceived exertion (RPE) threshold (PET) and neuromuscular fatigue threshold (NFT). Six trained male cyclists (30.2±5.7 years; 78.9±11.3 kg; 181±10 cm; 61.6±5.7 mlO2/kg/min) performed an incremental test for peak power output (PPO) and three predictive constant-load cycling tests at 88% (T88%), 94% (T94%) and 100% (T100%) of PPO under normoxia (FIO₂ = 20.93% FIO₂) and hypoxia (FIO₂ = $15\%O_2$). For each FIO₂ condition, slopes of RPE for legs (RPE-L), breathing (RPE-B) and overall (RPE-O) were used calculate PET for legs (PET-L), breathing (PET-B) and overall (PET-O) and with the muscle activity (EMG_{slopes}), NFT was established by the same protocol. Overall and differentiated PET were lower under hypoxia compared to normoxia (P < 0.05), whereas no significant difference was found for NFT between conditions (P > 0.05). Comparisons between PET modes within conditions revealed no significant differences (P > 0.05) in normoxia, while PET-L was significantly lower (P < 0.05) than PET-B and PET-O under hypoxia. In normoxia, PET modes were correlated to NFT (r=0.90) and highly related among each other (r=0.99; P<0.01) and under hypoxia, PET modes showed no significant relationship to NFT (P>0.05), but maintained its association among each other. Thus, we conclude that PET, in particularly PET-L, to be responsive to a decreased fraction of inspired O₂, but not NFT. These findings extend the use of overall and differentiated PET in moderated altitudes.

INTRODUCTION

Rating of perceived exertion (RPE) has been widely used to monitor and prescribe exercise intensity in clinical and sports settings exercise (American College of Sports Medicine, 2005; Borg, 1998) including several environmental conditions such as in altitude (Maresh *et al.*, 1993). This psychophysiological construct thought is indicated to correspond to a conscious/verbal manifestation of effort resulted from interpretations of many physiological and psychological signals integrated in the central nervous system (St Clair Gibson and Noakes, 2004; Noakes, 2008; Tucker, 2009). Although the mediators mechanisms of RPE are controversial (Noakes, 2011; Marcora, 2009), this parameter has shown to predict maximal oxygen uptake (Eston *et al.*, 2008) and exercise performance (Crewe *et al.*, 2008; Pires *et al.*, 2011).

Linear RPE increases (RPE_{slope}) during constant-load has been consistently shown during constant load exercise (Noakes, 2004; Nakamura et al., 2005; Eston et al., 2007). RPE_{slope} revealed to be related with intensity performed and time to exhaustion (Nakamura et al 2005, Fontes et al., 2010). Therefore, Nakamura and colleagues (2005a,b; 2009) proposed a method to estimate the critical power (CP) and the maximal steady state of oxygen consumption (VO₂), based on the RPE responses over 3-4 severe tests (i.e., above CP). This perceived exertion threshold (PET) was calculated from the theoretical intensity in which the RPE would not increase over time, from the estimate of the x intercept of the linear relationship between the exercise intensity (x axis) and the increase rate of the RPE (y axis) over the duration of the severe tests. PET has shown to be reliable aerobic index (Nakamura et al., 2009), similar and high correlated with critical power (r=0.87-0.98), ventilatory thresholds (VT₁ r=0.76; VT₂ r=0.72) and maximal oxygen consumption steady state intensity (r=0.92) (Nakamura et al., 2005a; 2005b; 2008; 2009). With similar protocol and linear regressions analysis, the neuromuscular fatigue threshold (NFT) has been proposed to estimate fatigueless intensity with muscle activity (EMG). NFT has been also related to ventilatory threshold (r=0.82; 0.92) (Matsumoto et al., 1991; Moritani et al., 1993) and rowing performance (r=0.96) (Maestu et al., 2006), although its validity as an aerobic index has been questioned (Maestu et al., 2006; Pavlat et al., 1995). More recently, we have compared PET and NFT estimations during same exhaustive bouts and showed no significant differences and higher relationship between them (r=0.78) (Fontes et al., 2010).

Thus, we suggested the close association of between the muscle recruitment and perception of effort during exhaustive exercise.

In addition to overall RPE (RPE-O), this parameter can be differentiated in breathing (RPE-B) (e.g., cardio-respiratory, chest) and legs (RPE-L) (e.g., local, muscular) components, which in turn are considered to be coupled to specific physiological mediators related to the chest and the working limbs, respectively (Robertson & Noble, 1997; Maresh *et al.*, 1993; Pandolf, 1982). Although differentiating RPE slopes could provide insightful information on the perceptual signal dominance during whole body dynamic exercise, limited information is available regarding these variables (Lima-Silva *et al.*, 2010). Here we investigated the predictions of PET by differentiated RPE which might bring a new possibility to estimate separately fatigueless intensity. This prediction could enhance PET's practical application, since exercise training adaptations could be easier monitored by this simpler method.

Hypoxic conditions has been shown to decreased exercise performance (Clark *et al.*, 2007; Amann *et al.*, 2007; Romer *et al.*, 2007) and aerobic capacity estimated by lactate threshold, muscle deoxygenation (Lorenz et al., 2006), ventilatory parameters (Hughson *et al.*, 1995) and critical power (Dekerle *et al.*, Epub ahead of print) despite the controversy about the physiological mechanisms explaining these responses (Amann and Kayser, 2009; Kayser, 2003). There are evidences that whole body and differentiated RPE in response to exhaustive exercise are also affected by the fraction of inspired oxygen (FIO₂) (Romer *et al.*, 2007) being RPE-B more commonly accentuated (Cibella *et al.*, 1999; Lane *et al.*, 1990), however it might be task dependent. In addition, skeletal muscle activity has also shown to be affected by hypoxia (Kayser *et al.*, 1994, Goodal *et al.*, 2010; Romer *et al.*, 2007), however, neither PET nor NFT have been investigated under this condition. Furthermore, the prediction of fatigueless intensity by a non-expensive method on decrease FIO₂ will extend its practical application to environments with similar characteristics such as in moderated altitude.

Thus, the purposes of this study were to verify the effects of hypoxia on overall and differentiated PET and NFT. We hypothesize that decreased FIO_2 would induce higher breathing rate and simultaneously cause higher increase rates of RPE modes, particularly to RPE-B, as well as EMG, thus reducing its predicted fatigueless intensity.

METHODS

Subjects

Six well trained male cyclists $(30.2 \pm 5.7 \text{ years}; 78.9 \pm 11.3 \text{ kg}; 181 \pm 10 \text{ cm}; 61.6 \pm 5.7 \text{ mlO}_2/\text{kg/min}; 8.3 \pm 2.2 \text{ training years}; 6.0 \pm 3.1 \text{ training hours per week}, 316 \pm 209 \text{ km per week})$ signed an informed consent and volunteered to take part in this study after being properly instructed about the purposes, procedures, risks and benefits of the investigation. Athletes were instructed to avoid exhaustive exercise 24h hour preceding all trials and to not ingest caffeinated substances neither alcoholic drinks during the experimental period. All subjects were well familiarized to the experimental procedures since they had already participated in previous studies using similar methods. In addition, they were used to complete exhaustive high intensity efforts during their day-by-day cycling training routine. The study protocol was approved by the local ethics committee and completed according to the declaration of Helsinki.

Experimental design

Subjects reported to the laboratory on seven occasions separated by at least 48 h during an 8-week period. Three exhaustive constant-load cycling bouts were performed under normoxia and hypoxia at different intensities based on peak power output reached in an incremental test. Cyclists completed the tests at approximately the same time of the day with ambient temperature ranging from 21 to 23°C and 60% of humidity. Differentiated ratings of perceived exertion and muscle activity were assessed during all tests. Performance (i.e., time to exhaustion), peak oxygen uptake and plasma lactate were collected at the end of each test. For each inspired oxygen condition, RPE scores were used to estimate the PET and EMG for NFT.

Predictive tests

During the predictive tests, three bouts were performed under nomoxic (FIO₂ = 20.93 %) and three under hypoxic (FIO₂ = 15 %) conditions. Exercise intensities were set at 88% (T88), 94% (T94) and 100% (T100) of PPO established by a previous performed incremental test (start 100W with 20W.min⁻¹; ~85 rpm; until exhaustion). A counterbalanced order among intensities and conditions was applied. Subjects initially rested five min baseline with the respective fraction of inspired oxygen followed by five min cycling warm up at 100W. Then,

after 2 min rest, the predictive tests started and were completed until exhaustion. Athletes completed all tests on their own bicycles attached to a cycling simulator (Computrainer Pro, Racer Mate, Seattle, USA) and kept the cadence at ~85 rpm. When cyclists were unable to maintain the proposed cadence for more than five seconds, exhaustion was taken to the nearest second. Athletes received strong verbal encouragement during all tests. Exercise intensity, performed duration and O_2 availability were blinded among the subjects during all study period.

Inspired O₂ concentration

The normobaric and decreased fraction of inspired oxygen condition (FIO₂ 15%) was controlled by an altitude simulator (AltiTrainer200, SMTEC, Switzerland) which offers air through a face mask. Four meters long tube was connected to a valve (2700 Two-way non-rebreathing valves T-ShapeTM, Kansas, USA) and attached to the mask. Subjects were blinded to the O₂ supply condition since tube connection to simulator was hidden and during the normoxic trials, a small fan was attached to the equipment to simulate the subject's ventilation and produce the similar noise from the hypoxic trials.

Ratings of perceived exertion

Ratings of perceived exertion for the legs (RPE-L), breathing (RPE-B) as well as overall RPE (RPE-O) was reported by using Borg's 6-20 scales (Borg, 1982). Subjects reported RPE scores every 30 seconds on a randomized order. Before each test standard instructions were provided on the RPE anchoring as follows: "number 7 represents unloaded cycling while number 19 indicates an exertion similar to exhaustive cycling, please, when asked, separately report your exertion for legs, breathing and overall to your whole body exertion". RPE scores were plotted against time in each predictive trial and the slopes from the linear regression were taken for overall (RPE-O_{slope}), legs (RPE-L_{slope}) and breathing (RPE-B_{slope}).

EMG recordings (EMG)

After shaving and cleaning the left vastus lateralis (VL) area with ethanol, two electrodes (Blue Sensor, Medicotest, Denmark) were taped 20 mm apart and parallel to the muscle fibers. VL electrodes were placed at two thirds of the distance between the anterior iliac crest and lateral border of patella. A telemetric EMG system (TeleMyo 2400T G2, Noraxon,

Arizona, USA) was used. Data were collected at 2000 Hz and band pass filtered at 20-500 Hz with the software MR-XP 1.07 Master Edition (Noraxon, Arizona, USA). EMG data was treated as five seconds root-mean-square (RMS) windows and normalize to the initial five seconds period of the respective test. EMG slopes for the VL (EMG_{slope}) were obtained for each trial by the regression angular coefficients found between EMG and time (DeVries *et al.*, 1982; Maestu *et al.*, 2006).

VO₂ and lactate

Ventilatory parameters were continuously monitored and averaged over eight respiratory cycles through all tests using a breath-by-breath gas analyzer (K4b2, COSMED, Rome, Italy). The system was calibrated according to the manufacturer's instructions before each test with a 15% O2 and 6% CO2 reference gas mixture and a 3 L syringe. Peak oxygen uptake during all trials and conditions was considered the highest value attained.

At the third and fifth minutes after exercise cessation 100 µl blood samples were collected from subject's ear lobes for posterior determination of plasma lactate concentration (YSI 2300 STAT PlusTM, Yellow Springs, USA), being the highest value [lac]p considered for analysis.

Perceived exertion (PET) and neuromuscular fatigue (NFT) thresholds

The slopes from overall and differentiated RPE, as well EMG from the predictive trials were respectively used for perceived exertion (PET) and neuromuscular fatigue threshold (NFT) determinations. Thus, individual regression lines relating RPE and EMG slopes to exercise intensities (in Watts) were estimated and the y-intercepts of these regressions lines indicating zero slopes for RPE and EMG were considered PET (Nakamura *et al.*, 2005, Fontes *et al.*, 2010) and NFT (DeVries *et al.*, 1982; Maestu *et al.*, 2006), respectively.

Statistics

Data were presented as median \pm semi-interquartile range (SIR). Nonparametric tests were employed for inferential analysis since Shapiro Wilk's test confirmed the non-normality of data set. Comparisons among fatigueless exercise intensity estimations and between O₂ availability conditions were performed by Friedman's test followed by Wilcoxon's test as *pos hoc* when pertinent. Relationships among fatigueless estimations were verified by Spearman's correlations. Statistical analysis was carried out using a statistical software package (SPSS for Windows, SPSS Inc., Chicago, USA) and in all cases statistical significance was set at P < 0.05.

RESULTS

Times to exhaustion (T_{EX}) under hypoxia (256.0 ± 28.0, 180.0 ± 22.1 and 136.5 ± 7.9 s) were significantly lower (P < 0.05) than those under normoxia (378.0 ± 29.5, 240.5 ± 31.0 and 176.5 ± 17.3 s for T88%, T94% and T100%, respectively) for all exercise intensities. Moreover, in both conditions T_{EX} at T88% were greater than those at T94% and T100% (P < 0.05), which also differed significantly (P < 0.05) from each other. Peak VO₂ varied from 57.7 to 61.6 mlO₂/kg/min under normoxia, and from 51.2 to 53.4 mlO₂/kg/min under hypoxia, being significant differences observed between conditions (P < 0.05) but not intensities (P > 0.05). In addition, no significant differences among intensities or conditions were found for plasma lactate, which ranged from 12.8 to 13.9, and 14.5 to 15.2 mmol/l for normoxia and hypoxia, respectively. Overall and differentiated RPE reached at the end of predictive bouts were similar irrespective of condition and intensities (P > 0.05).

Overall and differentiated RPE slopes as well as EMG slopes rose with increasing exercise intensities in both normoxia and hypoxia, being higher values observed in the latter condition (Table 6). Pairwise comparisons showed all RPE_{slope} modes to be significantly higher (P < 0.05) in hypoxia than in normoxia at T88% and T94%, but not (P > 0.05) at T100%. Moreover, again all RPE slopes at T100% were significantly higher (P < 0.05) than those at T88% in both conditions, and overall RPE slope at T100% under hypoxia was also greater (P < 0.05) than that at T94%. When comparing RPE slope modes in each intensity and condition, the only significant was found at T100% in hypoxia, with higher slope for RPE-O than for RPE-L (P < 0.05). Regarding EMG slopes, significant differences were observed between normoxic and hypoxic conditions for T88% and T94%, whereas values at T100% were greater than those the others in both conditions.

Table 6. Slopes of perceived exertion for legs (RPE-L_{slope}), breathing (RPE-B_{slope}) and overall (RPE-O_{slope}) and EMG (EMG_{slope}) during predictive bouts performed until exhaustion at 88% (T88%), 94% (T94%) and 100% (T100%) under normoxia and hypoxia.

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	Т8	8%	Т9	4%	T100%		
	Normoxia	Hypoxia	Normoxia	hypoxia	Normoxia	Hypoxia	
RPE-L _{slope} (units.min ⁻¹)	1.31 ± 0.08	$2.17 \pm 0.42*$	1.71 ± 0.14	$2.89 \pm 0.68*$	$2.99 \pm 1.15^{\#}$	$4.10 \pm 0.70^{\#}$	
$RPE-B_{slope}$ (units.min ⁻¹)	1.00 ± 0.09	$1.94 \pm 0.10^{*}$	1.69 ± 0.61	$3.91 \pm 0.71^*$	$3.63 \pm 1.34^{\#}$	$5.30 \pm 1.30^{\#}$	
RPE-O _{slope} (units.min ⁻¹)	1.14 ± 0.15	$1.90 \pm 0.37^{*}$	1.74 ± 0.16	3.10 ± 0.87*	$3.41 \pm 1.20^{\#}$	$4.90 \pm 0.33^{\# \pm \infty}$	
EMG (RMS%.min ⁻¹)	0.02 ± 0.04	$0.10 \pm 0.06*$	0.09 ± 0.05	$0.13 \pm 0.05*$	$0.21 \pm 0.09^{\#}$	$0.33 \pm 0.13^{\#}$	

n = 5 for T88% and n = 6 for T94% and T100%. *P < 0.05 from the same intensity in normoxia; $^{\#}$ P < 0.05 from T88% in the same condition; ‡ P < 0.05 from T94% under same condition; $^{\infty}$ P < 0.05 from RPE-L at same intensity and condition.

RPE slopes increased linearly as a function of exercise intensity, with R^2 ranging from 0.85 to 0.92 in normoxia and from 0.84 to 0.96 in hypoxia. For EMG slopes, R^2 ranged from 0.68 to 0.99 and from 0.70 to 0.99 for normoxia and hypoxia, respectively. PET and NFT determinations for a representative subject under both oxygen availability situations are shown in figure 1A-D.



Figure 8. Determination of perceived exertion threshold for legs (PET-L) (A), breathing (PET-B) (B) and overall (PET-O) (C) and neuromuscular fatigue threshold (NFT) (D) under normoxia and hypoxia. Data from a representative subject.

Overall and differentiated PET were lower under hypoxia compared to normoxia (P < 0.05), whereas no significant difference was found for NFT between conditions (P > 0.05). Comparisons between PET modes within conditions revealed no significant differences (P > 0.05) in normoxia, while PET-L was significantly lower (P < 0.05) than PET-B and PET-O under hypoxia. In neither condition NFT differed significantly (P > 0.05) from any PET (Figure 9).



Figure 9. Leg (PET-L), breathing (PET-B), and overall (PET-O) perceived exertion thresholds and neuromuscular fatigue threshold (NFT) under normoxia and hypoxia. *P < 0.05 from normoxia; $^{*}P < 0.05$ from PET-B and PET-O under hypoxia.

Relationships between fatigueless intensity estimations by the different parameters in each situation are presented in table 7. In addition, significant between condition relationships were found for PET-L (r = 0.90; P < 0.05), PET-B (r = 0.99; P < 0.01) and PET-O (r = 0.90; P < 0.05), but not NFT (r = 0.80; P > 0.05).

	ĕ		
	PET-B	PET-O	NFT
normoxia			
PET-L	0.99**	0.99**	0.90*
PET-B	-	0.99**	0.90*
PET-O	-	-	0.90*
hypoxia			
PET-L	0.90*	0.99**	0.70
PET-B	-	0.90*	0.60
PET-O	-	-	0.70
0.05 440 0.01			

Table 7. Relationships between perceived exertion threshold for legs (PET-L), breathing (PET-B), overall (PET-O) and neuromuscular fatigue threshold (NFT) under normoxia and hypoxia.

*P < 0.05; **P < 0.01

DISCUSSION

The aims of the present investigation were to verify the influence of decreased fraction of inspired oxygen on the fatigueless intensity estimates predicted by overall and differentiated RPE and EMG. Our main findings were that all PET were significantly lower under hypoxia and being PET-L reduced to a great extent compared to PET-B and PET-O. On the other hand, NFT was not affected by the experimental manipulation. Therefore, fatigueless intensity estimations by perceived exertion might be a responsive method for moderated altitude conditions.

Exercise performance under hypoxic exposure has been commonly shown to be reduced (Clark *et al.*, 2007; Amann *et al.*, 2007; Romer *et al.*, 2007). These decrements are normally accompanied by attenuation on aerobic capacity estimates, such as lactate threshold, ventilatory threshold (Hughson *et al.*, 1995) and critical power (Dekerle *et al.*, Epub ahead of print). The present study is the first to investigate the estimations of PET and NFT under hypoxic conditions. We showed that overall and differentiated PET were lowered (P<0.05) when fractions of inspired oxygen was decreased. The PET makes use of similar assumptions from critical power model, which the increase rate of RPE is associated with energy depletion of anaerobic sources (Nakamura *et al.*, 2009). Nakamura and colleagues (2009) suggested that this aspect would cause disturbances in cellular acid-basis and act as the main afferent signal source to RPE increase in severe exercise. Therefore, the decreased oxygen availability during the predictive tests might had increase this local disturbances and accentuate the rate of RPE increase, as shown by our data. However, the RPE formation is still controversial (Marcora, 2009), since different physiological responses such as cerebral oxygenation might also affect its responses (see Chapter 2).

When PET was estimated by differentiated RPE under normoxia, significant correlations (r=0.99) and similar intensity predictions were found to whole body PET. These results indicate that the rate of perceptions of effort among modes increased in parallel during all predictive trials under normoxia and may suggest their influence on each other. Additionally, our actual data corroborates with our previous findings (Fontes *et al.*, 2010) by estimating similar PET-O and NFT. On that study, we concluded that the slope of muscle recruitment might be an important response to PET predictions. Furthermore, early study (Chapter 2) showed that these

muscular responses are also associated to the perception of effort for legs and breathing, which can explain the close intensity estimations by differentiated PET and NFT.

If on one side our data showed that overall and differentiated PET to be responsive to the effects of moderated hypoxia, on the other, NFT did not, even though EMG slopes also increased under this condition. Both methods are derived from slopes of linear regression analysis which extremity values play a greater role on its inclination. Then, when discrepancies on the magnitude of changes of these points are found, slopes might alter significantly and not show the effect of the experimental comparison. PET showed similar determinations slopes as reflected by similar magnitude changes influenced by hypoxia in all workloads. However, when having a closer look to the NFT determination slopes, EMG_{slope} under hypoxia at T88% increased 400% when compared to normoxia. Given the higher aerobic nature of this workload performed (T88%), hypoxia seems to exert higher influence on the rate of muscle activity showed by our data, thus consequently, prediction slope were attenuated and the effects of hypoxia not showed on the predicted fatigueless intensity. Furthermore, we have also shown that NFT to not be responsive to caffeine supplementation (Gonçalves *et al.*, 2010) even though increases on EMG slopes were found, thus similar discrepancies among the changes on the extremities values of regression analysis for its determination might explain this results.

Evidences have shown the influence of lower FIO₂ and exercise intensities on whole body and differentiated RPE (Romer *et al.*, 2007; Maresh *et al.*, 1993), in particular for breathing RPE (Cibella et al., 1999; Lane et al., 1990). Since specific physiological responses have been considered to be tied to perceived exertions for legs and breathing (Robertson and Noble 1997; Pandolf, 1982), we expected that the hyperventilation usually showed under hypoxia (Cibella *et al.*, 1999; Lane *et al.*, 1990) would induce higher slope rates for RPE-B and decrease PET estimations. However, our data showed that the more affected PET estimation was for legs and not for breathing nor overall. These findings may indicate the role of peripheral effort perceptions on the estimation of fatigueless exercise intensity. In agreement with our results, Shepard and colleagues (1992) suggested that exercise intensity prescription might be more appropriated by peripheral effort sensations than for respiratory and overall RPE under hypoxia. However, future studies are needed to better clarify this issue. Based on our results, we propose two main issues to improve PET's practical applications. Due to the simplistic application and responsiveness to lower levels of oxygen availability, we suggest the extension of PET under moderated altitudes to facilitate the prediction of training intensities on this condition. The second suggestion is the possibility to estimated peripheral and respiratory aerobic capacity by the ratings of perceived exertion. Since RPE showed to be coupled to muscular activity, specific training adaptations might be also monitored when applying this method separately.

It is important to emphasize that RPE scale requires familiarization. Subjects that may have not experienced higher levels of exercise intolerance might not inform reliable scores. Thus, we suggest the use of RPE on different exercise sessions with well described instructions and scale anchoring to minimize erroneous analysis. In addition, the quantity (~3-4) of exhaustive tests performed to estimate the fatigueless intensity by PET might limit its practical application. Nevertheless, reliable estimations of PET by non-exhaustive bouts have been already presented (Nakamura *et al.*, 2008).

CONCLUSIONS

Data from the present study suggest PET and particularly PET-L to be more responsive to a decreased fraction of inspired O_2 compared to NFT. These results can be explained by the differential effects of hypoxia on the slopes of linear regressions of RPE and EMG slopes against power output.

4 CONSIDERAÇÕES FINAIS

No decorrer de toda evolução da espécie, o ser humano tem convivido com enorme dispêndio energético e quase que invariavelmente associado a constante necessidade de exercitar-se, seja caçando durante a pré-história, plantando na idade média ou correndo para pegar um taxi nos dias de hoje. Com o avanço tecnológico dos últimos 50 anos, muitas dessas necessidades diminuíram drasticamente, já que diferentes aparelhos eletrônicos ou meios de transporte parecem substituir mais e mais nossos esforços. Diante dessas observações, surge a questão: a evolução nos direciona para um caminho que não estamos preparados?

De qualquer forma, nossa situação atual remete a uma evolução no qual exercitar-se parece ser uma necessidade fisiológica e talvez também psicológica. Atualmente, se para alguns, a prática de exercícios físicos é apenas uma forma de manter-se saudável, bonito, ou mesmo campeão, para outros, seu entendimento mais aprofundado pode fazer com que mais pessoas pratiquem mais exercícios de forma adequada e prazerosa, para assim alcançarem seus objetivos internos, seja lá qual forem eles. Dentro dos principais fatores que inferem na realização da atividade física, o controle da intensidade pode ser considerado um dos mais importantes. Constantemente associado ao objetivo a ser alcançado, a intensidade do exercício pode tornar o ato prazeroso e saudável, ou ainda, desconfortável ou lesivo. Diante de tal importância, muitos estudiosos procuraram desenvolver diferentes métodos de entendimento e aplicação desse controle de intensidade. A grande maioria desses métodos utiliza-se de recursos e equipamentos extremamente custosos nos quais normalmente acompanham um considerável entendimento técnico desses aparelhos e procedimentos. No entanto, ao enumerarmos os métodos que realmente contribuíram para alguma aplicação prática, pouquíssimos poderão ser descritos, mesmo que esses tenham dominado amplamente os principais periódicos científicos nos últimos 100 anos. Acredita-se ainda que o grande problema associado a essas metodologias pouco práticas indica que essas buscaram o entendimento do exercício físico de forma isolada do comportamento da fisiologia e acabaram deixando de lado aspectos associados ao cérebro/mente.

Atualmente, Noakes (2011) apresenta o problema associado aos modelos tradicionais de fadiga, e indica que esses modelos nunca consideraram a interação óbvia do cérebro como o controlador de todas nossas tarefas motoras, assim como a manutenção da

homeostase. Normalmente, nosso comportamento motor está intimamente relacionado a uma resposta consciente de nossas ações ou intenções. Por outro lado, um controle inconsciente do sistema autônomo é apresentado no sentido de regulação e manutenção da homeostase. Quando um estímulo gera uma perturbação nesse controle, ajustes conscientes da tarefa motora e mesmo inconscientes sobre a regulação autonômica são sempre gerenciados pelo cérebro. É vastamente proposto na literatura que esse controle consciente da tarefa motora reflete sobre a percepção subjetiva do esforço (PSE). Normalmente as respostas dessa percepção são adquiridas por métodos extremamente simples e subjetivos, como uma escala analógica. No entanto, apesar de PSE estar sendo estudada a mais de 150 anos, sua formação ainda não é clara. Estudos apontam a estrita relação entre PSE e limites do ser humano durante o exercício. Dessa forma, acredita-se que trabalhos que venham a auxiliar no entendimento da formação de PSE auxiliarão o uso mais adequado desse método para predizer capacidades físicas e controlar intensidade. Assim, há a possibilidade de que a prática de exercícios físicos se faça de uma maneira mais eficaz, prazerosa e saudável.

O presente trabalho buscou entender melhor a formação de PSE através da aplicação de protocolos exaustivos e manipulação da fração de oxigênio inspirado para instalação da fadiga. Durante o primeiro trabalho (Capitulo 1), sujeitos saudáveis foram submetidos a esforços em cargas constantes e exaustivas no qual a PSE e o recrutamento adicional de fibras musculares (EMG) foram monitorados. Os resultados apontaram uma estreita relação entre essas variáveis e quando estimou o limiar de esforço percebido (LEP) e o limiar de fadiga neuromuscular (LFN), apresentou estimativas de intensidades similares e forte correlação entre os métodos. Nesse trabalho, indicamos a importante associação entre PSE e EMG e sugerimos que PSE pode ser considerada como um simples e válido indicador indireto de recrutamento muscular.

Diante da necessidade da investigação da participação do sistema nervoso central na formação de PSE, foi elaborado um segundo estudo. Além disso, no sentido de expandir os achados para outras realidades ambientais e entender os possíveis mecanismos sobre PSE, o objetivo foi explorado também sobre situações com diminuídas ofertas de oxigênio. Os achados desse estudo indicaram não apenas a significativa relação de PSE com EMG, mas também com a oxigenação cerebral. Além disso, PSE foi novamente associada ao desempenho em normóxia, hipóxia e à própria oxigenação cerebral. Os resultados apontam para uma

importante interação das respostas periféricas e centrais na formação de PSE, assim como o controle do desempenho.

Se por um lado, os dois primeiros trabalhos desse documento buscaram os mecanismos de formação de PSE e apresentaram uma aproximação mais teórica sobre esse fenômeno, o terceiro, verificou a possibilidade de ampliar a aplicação prática dessa variável. Foram estimados os limiares de esforço percebidos de formas diferenciadas, para o corpo como um todo (LEPgeral), respiração (LEPrespiração) e local (LEPlocal) além de serem comparados ao limiar de fadiga neuromuscular. Estes não apresentaram diferenças significativas, portanto, foi possível propor a aplicação desses métodos por apresentarem uma maneira mais simples para monitorar adaptações diferenciadas ao treinamento. Além disso, esse trabalho apresentou a possibilidade de utilizar-se LEP em locais com altitudes moderadas.

Perspectivas futuras

Com o avanço tecnológico, é percebido um significativo aumento da procura por temas relacionados ao sistema nervoso central, quando associado ao exercício. Assim, a participação do controle cerebral durante o exercício começa a tomar o caminho tão pouco explorado nas ultimas décadas. Nesse sentido, em colaboração com o Grupo de Estudos em Biologia Integrativa do Exercício (UFRN), foi desenvolvido um trabalho no qual verificamos os efeitos da estimulação elétrica transcraniana por corrente contínua direcionada ao córtex insular esquerdo. Demonstramos que apenas atletas sofreram aumento da atividade parassimpática após aplicação da estimulação e especulamos uma possível adaptação local dessa área por efeitos do controle exercido durante o exercício físico crônico. Assim, como já proposto anteriormente, o córtex insular demonstrou seu controle sobre a regulação autonômica. Vale ressaltar que essa área exerce importantes ações sobre o controle estável da homeostase, como modulação da freqüência cardíaca, pressão arterial e retenção de sal.

Em um segundo trabalho, realizado também em colaboração com o mesmo grupo nordestino, verificamos os efeitos da mesma estimulação transcraniana sobre o córtex insular e verificamos o desempenho e possível modulação da PSE (Okano *et al* em elaboração). No entanto, parece que além de estar associado a um controle inconsciente da manutenção da homeostase durante o exercício, os achados preliminares desse estudo mostram que essa área pode estar também associada a um suposto controle consciente. Dessa forma, sugere-se a associação de novas tecnologias com metodologias inovadoras para buscar um maior entendimento do controle cerebral sobre o exercício. No entanto, o desafio pela aplicação prática desses novos e futuros estudos permanece em aberto.

Apesar de ter sido demonstrado a associação da oxigenação do córtex préfrontal com o desempenho e a PSE, outras áreas cerebrais superficiais ou mesmo profundas, como o córtex insular, podem também participarem dessa regulação central do exercício. Assim, em colaboração com o grupo de pesquisa "Central Governor Group" e sob orientação do Dr Noakes, Daniel Bodnariuc Fontes e Alvaro Fontes aceitaram o desafio e desenvolveram um protótipo para viabilizar a realização da análise funcional do cérebro durante o exercício do ciclismo. Esse trabalho piloto apresentou de forma original a possibilidade de analisar a atividade cerebral e associá-la a percepção de esforço. Os achados desse projeto paralelo serão apresentados no 16º Annual European College of Sports Science na cidade de Liverpool, 2011. Com essa metodologia desenvolvida, espera-se que novas fronteiras venham a serem abertas para o maior entendimento da regulação cerebral sobre o exercício, por meios conscientes ou inconscientes. Assim, parece que nossos corpos estão mais próximos das nossas mentes do que imaginamos.

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Apêndices

- 1 Comitê de Ética
- 2 Consentimento informado
- 3 Informações gerais aos sujeitos
- 4 Ficha de avaliação
- 5 Escalas utilizadas
- 6 Check list de procedimentos





Health Sciences Faculty Research Ethics Committee Room E52-24 Groote Schuur Hospital Old Main Building Observatory 7925 Telephone [021] 406 6626 • Facsimile [021] 406 6411 e-mail: shuretta.thomas@uct.ac.za

17 April 2009

REC REF: 100/2009

Mr E Fontes c/o Prof T Noakes Human Biology Sport Science Institute

Dear Mr Fontes

PROJECT TITLE: EFFECT OF ACUTE HYPOXYA ON THE PERCEIVED EXERTION THRESHOLD, CRITICAL POWER AND ELECTROMYOGRAPHIC SIGNAL.

Thank you for submitting your study to the Research Ethics Committee for review.

It is a pleasure to inform you that the Ethics Committee has formally approved the above-mentioned study.

Approval is granted for one year till the 25th April 2010.

Please submit an annual progress report if the research continues beyond the expiry date. Please submit a brief summary of findings if you complete the study within the approval period so that we can close our file.

Please note that the latest version of the Helsinki Declaration is 2008. Please check the following sentence in the information sheet (page2) as it sounds confusing: "Blood assistance be analysed in a laboratory..."

Please note that the ongoing ethical conduct of the study remains the responsibility of the principal investigator.

Please quote the REC. REF in all your correspondence.

S Thomas

Yours sincerely		Z
	(/

PROFESSOR M BLOCKMAN CHAIRPERSON, HSF HUMAN ETHICS

Federal Wide Assurance Number: FWA00001637. Institutional Review Board (IRB) number: IRB00001938

This serves to confirm that the University of Cape Town Research Ethics Committee complies to the Ethics Standards for Clinical Research with a new drug in patients, based on the Medical Research Council (MRC SA), Food and Drug Administration (FDA-USA), International Convention on Harmonisation Good Clinical Practice (ICH GCP) and Declaration of Helsinki guidelines.

The Research Ethics Committee granting this approval is in compliance with the ICH Harmonised Tripartite Guidelines E6: Note for Guidance on Good Clinical Practice (CPMP/ICH/135/95) and FDA Code Federal Regulation Part 50, 56 and 312.

λ.

S Thomas





Health Sciences Faculty Research Ethics Committee Room E52-24 Groote Schuur Hospital Old Main Building Observatory 7925 Telephone [021] 406 6626 • Facsimile [021] 406 6411 e-mail: shuretta.thomas@uct.ac.za

25 August 2009

REC REF: 100/2009

Mr E Fontes Research Unit for Exercise Science & Sports Medicine Department of Human Biology

Dear Mr Fontes

PROJECT TITLES: EFFECT OF ACUTE HYPOXYA ON THE PERCEIVED EXERTION THRESHOLD, CRITICAL POWER AND ELECTROMYOGRAPHIC SIGNAL.

Thank you for your letter to the Research Ethics Committee dated 21st August 2009.

It is a pleasure to inform you that the Ethics Committee has **granted approval** for the amendment to use the new hypoxia simulator (AltiTrainer 200, Switzerland) and to conduct testing (Near Infrared Spectroscopy) at the Cape Universities Brain Imaging Centre at Tygerberg.

Should the latter investigations require additional transport costs for participants this needs to be reflected in the informed consent forms, plus whether these costs will be re-imbursed.

Please note that the ongoing ethical conduct of the study remains the responsibility of the principal investigator.

Please quote the REC. REF in all your correspondence.

Yours sincerely

PROFESSOR M BLOCKMAN CHAIRPERSON, HSF HUMAN ETHICS

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EFFECT OF ACUTE HYPOXYA ON THE PERCEIVED EXERTION THRESHOLD, CRITICAL POWER AND ELECTROMYOGRAPHIC SIGNAL

Dear Subject,

Thank you for your interest in participating in the above mentioned study.

Possible risks to the subjects

- The methods used during this study pose no inherent risk to you, other than the risks of performing exercise.
- Approximately 25 µL of blood (one drop) will be collected from the ear lobe at the end of each trial. The risk of drawing blood includes: infection, bruising, physical pain, mental discomfort and possible injury to a nerve or vessel. These risks or adverse effects are rare and will be minimised by the use of trained phlebotomists, and the use of sterile technique and disposable, single-use materials. Blood assistance be analysed in a laboratory for lactate and glucose concentration, after which it will appropriately be discarded.

Should any untoward events occur, effective treatment will be available from on-site medical care. Thus, any potential risks will be minimized. Finally, the University of Cape Town has a no-fault insurance or public liability cover should some unforeseen event occur whilst you are participating in this study.

The study will be performed in accordance with the principles of the Declaration of Helsinki, ICH Good Clinical Practice (GCP) and the laws of South Africa.

Benefits

We will inform you of the significance of our findings, once the results have been analysed.

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We will also provide you with a comprehensive assessment of your performance tests at both sea level and moderate altitude.

Statement of understanding and consent

I confirm that the exact procedures and techniques, and possible complications of the above tests have been thoroughly explained to me. I am free to withdraw from the study at any time, should I choose to do so. I understand that I may ask questions at any time during the testing procedure. I have been informed that the personal information required by the researchers, and derived from the testing procedure, will remain strictly confidential and will only be revealed as a number in statistical analysis.

I have carefully read this form and understand the nature, purpose and procedures of this study. I agree to participate in this research project of the MRC / UCT Research Unit for Exercise Science and Sports Medicine.

Name of volunteer:
Signature:
Name of investigator:
Signature:
Date:

Principal investigator: Eduardo Bodnariuc Fontes

UCT/MRC Research Unit for Exercise Science and Sports Medicine, PO Box 155, Newlands 7725.

Tel: 021-6504569

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Supervisor: Timothy D. Noakes UCT/MRC Research Unit for Exercise Science and Sports Medicine, PO Box 155, Newlands 7725. Tel: 021-6504569

Research and Ethics Committee of the Health Sciences Faculty, University of Cape Town Prof. M. Blockman Tel: 021-4066492





Research Project: EFFECT OF ACUTE HYPOXYA ON THE PERCEIVED EXERTION THRESHOLD, CRITICAL POWER AND ELECTROMYOGRAPHIC SIGNAL

General information for subjects

- 1 purpose of the study
- 2 responsible
- 3 my details
- 4 Tim's details
- 5 information sheet
- 6 funding
- 7 ethical aproval
- 8 data for research ONLY
- 9 schedule
- 10 recommendations:
 - training
 - eating
 - drinking
 - importance of serious attitude
- 11 drop out
- 12 doping
- 13 Thanks very much!!!





Research Project: EFFECT OF ACUTE HYPOXYA ON THE PERCEIVED EXERTION THRESHOLD, CRITICAL POWER AND ELECTROMYOGRAPHIC SIGNAL

subject					session date		time	
workload condition	vorkload temperature				VO2 file time to exl	haustion		
RPE							L	
	0.5	1	1.5	2	2.5	3	3.5	4
legs								
breathing								
overall								
* ^{3 0} - 4	4.5	5	5.5	6	6.5	7	7.5	8
legs								
breathing								
overall								
	8.5	9.0	9.5	10	10.5	11	11.5	12
legs								
breathing								
overall		1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1						
	12.5	13	13.5	14	14.5	15	15.5	16
legs								
breathing								
overall								

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RPE RECOVERY

	1	2	3	4	5
legs					
breathing					
overall					

obs

BREATHING

6

8

10

14

- 7 Very, very light
- 9 Very light
- 11 Fairly light
- 12
- 13 Somewhat hard
- 15 Hard
- 16
- 17 Very hard
- 18
- 19 Very, very hard
- 20

PET HYPOXIA

ANTES DOS SUJEITOS CHEGAREM

- Ligar o K4 40 minutos antes para aquecer
 - 1. Lactato: lancetas, luvas, algodão em pedaços pequenos, capilares, eppendorfs devidamente etiquetados, caixa com gelo para armazenar, álcool para limpeza.
 - 2. EMG: giletes, álcool, papel, fita adesiva, setup do sujeito no programa antes dele chegar.
 - 3. Borg: escalas
 - 4. VO2: máscaras, fixação do K4 na mesa
 - 5. Freqüência Cardíaca: relógio e belt
 - 6. Computrainer: verificar pneus, colocar a bicicleta na posicao, calibrar.
 - 7. Fichas: deixar as fichas para os sujeitos preparadas
 - 8. Calibrar o K4.

QUANDO OS SUJEITOS CHEGAREM

- 1. Colocar cinta do HR e testar.
- 2. Raspar vasto lateral e reto femoral e colocar os eletrodos. Testa-los.
- 3. Colocar mascara do VO2 e testar
- 4. Ligar Relógio, deixar o sujeito deitado por 5 minutos.
- 5. Desligar relógio.
- 6. Posicionar sujeito na bike
- 7. Realizar aquecimento
- 8. Calibrar bike novamente e colocar carga a ser realizada no teste.
- 9. Explicar procedimentos
- 10. Ligar VO2
- 11. Ligar EMG
- 12. Ligar relógio e cronômetro extra simultaneamente no momento de início do teste

DURANTE O TESTE:

- Coletar Borg (local, peito e geral)

FIM DO TESTE

- 1. LAP no relógio e cronômetro para marcar o descanso
- 2. Salvar arquivo de EMG
- 3. Lactato e percepção de fadiga à 1min, 3min e 5min do término.
- 4. Salvar VO2 e desligar relógio HR
- 5. Desligar cronômetro
- 6. Retirar a mascara de VO2 e eletrodos da EMG dos sujeitos
- 7. Retirar relógio e cinta do HR dos sujeitos
- 8. Desligar equipamentos se não tiver outro teste na sequência.
- Armazenar o lactato, passar dados do Suuntor para o computador, passar tempo de exaustao e Borg para excel.

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