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# original article

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# Oxygen consumption and heart rate obtained in a ramp protocol are equivalent during exercise session of rectangular loading at ventilatory thresholds for athletes

## *Consumo de oxigênio e frequência cardíaca obtidos em um protocolo de rampa são equivalentes durante sessão de exercícios de carga retangular nos limiares ventilatórios para atletas*

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**Abstract** – Training near or at ventilatory threshold (VT) is an adequate stimulus to improve the thresholds for sedentary subjects, but a higher intensity is necessary for conditioned subjects. The choice of cardiopulmonary exercise testing (CPx) protocol has an influence on VTs identification and can reduce their reliability for exercise prescription. This study tested if  $\dot{VO}_2$  and heart rate (HR) corresponding to first (VT1) and second ventilatory threshold (VT2) determined during a ramp protocol were equivalent to those observed in rectangular load exercises at the same intensity in runners elite athletes (EA) and non-athletes (NA). Eighteen health subjects were divided into two groups: EA (n = 9,  $\dot{VO}_{2max}$  68.6 mL·kg<sup>-1</sup>·min<sup>-1</sup>) and NA (n = 9,  $\dot{VO}_{2max}$  47.2 mL·kg<sup>-1</sup>·min<sup>-1</sup>). They performed CPx and 48h and 96h later, a continuous running lasting 1 h for VT1 and until exhaustion for VT2. The results showed that EA at VT1 session, presented delta differences for  $\dot{VO}_2$  (+9.1%, p = 0.125) vs. NA (+20.5%, p = 0.012). The Bland-Altman plots for VT1 presented biases of (4.4 ± 6.9) and (5.5 ± 5.6 mLO<sub>2</sub>·kg<sup>-1</sup>·min<sup>-1</sup>) and (4.9 ± 4.2 bpm). The ramp protocol used in this study was inappropriate for NA because it underestimates the values of  $\dot{VO}_2$  and HR at VT1 found in the rectangular load exercise. The HR showed good agreement at VT2 with CPx and may be a good parameter for controlling exercise intensity.

Key words: Exercise testing, Continuous running, Training prescription, Runners, Elite athletes

**Resumo** – O treinamento no limiar ventilatório (LV) é um estímulo adequado para melhorar os limiares em indivíduos sedentários, entretanto uma maior intensidade é necessária para indivíduos condicionados. A escolha do protocolo de teste de exercício cardiopulmonar (CPx) tem influência na identificação dos LV e pode reduzir sua confiabilidade na prescrição do exercício. Este estudo testou se o  $VO_2$  e a frequência cardíaca (FC) correspondentes ao primeiro (LV1) e segundo limiar ventilatório (LV2) determinados durante um protocolo de rampa foram equivalentes àqueles observados em exercícios de carga retangular nas mesmas intensidades em atletas corredores de elite (AE) e não atletas (NA). Dezoito homens saudáveis foram divididos em dois grupos: AE (n = 9,  $VO_{max}$  68,6 mL·kg<sup>-1</sup>·min<sup>-1</sup>) e NA (n = 9,  $VO_{2max}$  47,2 mL·kg<sup>-1</sup>·min<sup>-1</sup>). Eles realizaram CPx e 48h e 96h depois, uma corrida contínua com duração de 1 h para o LV1 e até a exaustão para o LV2. O grupo AE na sessão LV1, apresentou diferenças de delta para  $VO_2$  (+ 9,1%, p = 0,125) vs. NA (+ 20,5%, p = 0,012). Bland–Altman para LV1 apresentaram vieses de (4,4 ± 6,9) e (5,5 ± 5,6 mLO<sub>2</sub> · kg<sup>-1</sup> · min<sup>-1</sup>) mIO<sub>2</sub> · kg<sup>-1</sup> · min<sup>-1</sup> e (4,9 ± 4,2) bpm. O protocolo de rampa utilizado foi inadequado para NA pois subestima os valores de  $VO_2$  e pode ser um bom parâmetro para controlar a intensidade do exercício.

Palavras-chave: Teste de esforço, Corrida contínua, Prescrição de treinamento, Corredores, Atletas de elite

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#### INTRODUCTION

The cardiopulmonary exercise test (CPx) is the gold standard when determining maximal oxygen uptake ( $\dot{VO}_{2max}$ ) and in the prognosis and diagnosis of cardiopulmonary disease<sup>1</sup>. Individuals with different levels of physical performance like athletes and non-athletes perform CPx tests to improve training prescriptions obtained from  $\dot{VO}_{2max}$  fractions estimated by ventilatory threshold (VT), as submaximal control parameters during prolonged effort<sup>2,3</sup>. Coaches', athletes' and exercise physiologists' interest in determining and using VT is due to its strong correlation with lactate threshold and because it is a better performance prediction parameter than  $\dot{VO}_{2max}^{-1-4}$ .

VT intensity in relation to running speed, %VO<sub>2max</sub> and heart rate (HR) are all used to control intensity. However, the choice of ergometer and protocol used in CPx can affect the identification of the intensity associated with VTs and thereby reduce the reliability and reproducibility<sup>5</sup>. Ramp protocols allow individualisation of the test, more uniform haemodynamic responses and gas exchange and better prognosis, diagnosis and measurement of ventilatory thresholds than traditional staggered protocols<sup>6</sup>. However, the short duration of load changes in the ramp protocol may not show good reproducibility for prescription load on long-lasting exercise.

CPx performed on a cycle ergometer with smoothed load increments of between 20 and 50 W·min<sup>-1</sup> has shown reliable first ventilatory threshold (VT1) values and good VT1 and second ventilatory threshold (VT2) reproducibility in testing and retesting in individuals at different fitness levels<sup>5-7</sup>. However, protocols with large or uneven load increments (increments > 50 to 100 W·min<sup>-1</sup>) lead to a delay in  $O_2$  intake response in relation to the cycle ergometer workload and may overestimate  $\dot{VO}_{2max}$ on the treadmill<sup>5</sup>. A delay of 45 seconds or more in VT1 identification relating to workload was found in Davis et al.<sup>5</sup> therefore, these authors suggest caution when relating these values to VO<sub>2</sub> or HR for exercise prescription or performance evaluation<sup>5,6,8</sup>. Ramp protocols offer a means of overcoming the limitations of standard incremental protocols and, when the goal is to optimise exercise prescription, it is important to consider the difference between the values obtained in CPx and in the exercise session and whether this difference represents just a random fluctuation or a real change that requires the training load to be changed<sup>9</sup>.

It is known that athletes and non-athletes show stability in cardiorespiratory variables during an exercise session of constant load at the intensity of the VT1<sup>10</sup>. However, it is still unclear whether non-athletes present the same magnitude of response of the exercise prescription variables such as  $\dot{VO}_2$  and heart rate when compared between CPx and the constant loading session. It is possible that athletes present lower metabolic stress to sustain the load at the threshold due to better running economy<sup>11</sup>, lower systolic volume<sup>12</sup>. When compared to non-athletes was demonstrate previously that training at an intensity near or at ventilatory threshold is an adequate training stimulus for improving the thresholds for sedentary subjects, but a higher intensity may be necessary for conditioned subjects<sup>13</sup>. Therefore, the aim of this study was to test whether the ventilatory and metabolic variables corresponding to VT1 and VT2 determined during the ramp protocol are equivalent to those observed in a constant load exercise at the same intensity in both elite athletes and non-athletes. We hypothesized that subjects with higher fitness level (athletes) will present more equivalence at metabolic and ventilatory variables during constant load exercise than subjects with lower fitness level (non-athletes).

#### METHOD

#### **Participants**

A total of 18 subjects at different levels of cardiopulmonary performance, who were street runners with at least 1 year of experience, were evaluated and (characteristics of subjects are shown in Table 1). The subjects were selected by convenience and divided into two groups: 9 elite athletes (EA) that were highly trained and participated in races of 5 and 10 km, and 9 non-athletes (NA) classified a recreationally runner. A sample size calculation was used in accordance with the expected changes in  $\dot{VO}_{2max}$ , with SD of 8 mL·kg<sup>-1</sup>·min<sup>-1</sup> between groups(6)<sup>14</sup> using independent t test with  $(z\alpha = 1.96 \text{ and } z\beta = 0.84)$ . The volunteers were informed to abstained drink coffee, alcohol and exhaustive exercise almost 24h before visit at lab. At the first visit, they came at morning between 9 and 11h and performing anthropometry and cardiopulmonary exercise testing (CPx). The second and third visit, after 48h they run at first and second VT speed. The procedures were performed in accordance with local resolution no. 466, December 12, 2012, and were approved by the Federal University of Espírito Santo (UFES) ethics committee under nº. 261.897 of 02/05/2013. All subjects were informed about and familiarised with the experimental procedures, risks and benefits and signed terms of written consent<sup>15</sup>.

#### Procedures

The subjects visited the laboratory on three occasions separated by a minimum of 48 hours. Anthropometric and CPx were performed on the first visit, a 1-hour exercise session at VT1 speed was performed on the second visit and an exercise session at a speed of VT2, to volitional exhaustion, was performed on the third visit. HR and  $\dot{VO}_2$  was monitored in all visits. In addition, subjects were instructed to avoid strenuous exercise in the 24 hours preceding the laboratory visit. Subjects with cardiometabolic or uncontrolled musculoskeletal diseases that would prevent them from performing the tests were excluded. One EA group subject was excluded from the analysis due to technical problems with the heart rate monitor.

#### Anthropometrics

Body weight and height were measured using a digital scale with a onemillimeter precision stadiometer with a maximum capacity of 201kg and a sensitivity of 50 grams (Marte Científica, L200, São Paulo). It is possible to calculate the Body Mass Index (BMI). The anthropometric measurements were performed by a same experienced evaluator who used a scientific plicometer with a sensitivity of 0.1mm and reading range of 85mm (Mitutoyo / Cescorf, RS), including 7 skinfolds (triceps, subscapular, pectoral, supra-iliac, abdominal and thigh). For perimetry, was used a 2 m of flexible steel with resolution in mm (Cescorf, RS).

#### Cardiopulmonary Exercise Testing

Initially, the subjects remained for five minutes at rest in supine position. The 12-lead resting ECG (MICROMED, Brasília, Brazil) was performed to detect any disturbance that contraindicated maximum effort. Electrocardiographic recording during pre-exertion (standing on the treadmill) and exertion (simultaneously with CPx) was performed using three simultaneous leads (MC5, D2M and V2M).

The CPx was performed on a motorised treadmill (Inbrasport Super ATL, Porto Alegre, Brazil) with fixed 1% slope, following a ramp protocol with an estimated test duration of 10 to 12 minutes. The speed was gradually increased until exhaustion. The EA group started with a speed of 8 km·h<sup>-1</sup>, and the NA group commenced at a speed of 5 km·h<sup>-1</sup>, with an increase of 0.7 to 1 km·h<sup>-1</sup> every minute. A metabolic gas analyser (Cortex Metalyzer 3B, Germany) was used, with breath by breath collection and calibration with ambient and known gases (11.97% O<sub>2</sub> and 4.95% CO<sub>2</sub>). The volume was calibrated with a 3-L syringe. The temperature was controlled between 22 and 25°C. The test was conducted by a exercise physiologist and monitored by a cardiologist. At least three of the following criteria were considered in recognising the maximum test: a) volitional exhaustion; b) achieving HR  $\geq$  90% of the predicted maximum (220-age); c) respiratory exchange ratio (RER) of 1.05<sup>16</sup> or above; and d)  $\dot{VO}_{2max}$  plateau<sup>1,17</sup>. The maximum velocity (Vmax) was determined to be the rate at which the  $\dot{VO}_{2max}$  was identified.

#### Ventilatory Thresholds Identification

Three evaluators experienced in exercise physiology analysed the criteria of ventilatory thresholds blindly and independently. The intraclass correlation coefficient was used with values varying between (0.89 – 0.97). Agreement values between at two evaluators were considered. VT1 was identified by different criteria: 1- at the lowest point followed by an exponential increase in ventilatory oxygen equivalent ( $\dot{V}E/\dot{V}O_2$ ) without an increase in the ventilatory carbon dioxide equivalent ( $\dot{V}E/\dot{V}CO_2$ ); 2- V-slope method, indicating the point of intersection with loss of  $\dot{V}CO_2/\dot{V}O_2$  linearity; and 3-from abrupt increases in  $\dot{V}E$  and end-tidal oxygen tension ( $P_{\rm ET}O_2$ )<sup>18</sup>. VT2 was considered the point of the increase  $\dot{V}E/\dot{V}CO_2$  level, and beyond the point of gradual decrease of end-tidal carbon dioxide tension ( $P_{\rm ET}CO_2$ )<sup>18-19</sup>.

#### **Exercise Sessions**

The exercise sessions was made between 48h and 96h after CPx. The sessions commenced whit 5-minute warm-up, at an intensity of 20% below

the VT1 speed achieved in the CPx and free static stretches. The volunteers then remained standing for 2 minutes on the treadmill after the equipment calibration procedure, adjustments to the treadmill, mask and belt POLAR T31- CODED (Polar, Kempele, Finland) were performed. The stipulated speed was then adjusted manually. After 10 to 15 seconds of stabilisation, the subjects ran with continuous measurement of expired gases and a maximum duration of 1 hour in VT1. Forty-eight hours thereafter, the same procedures were performed in VT2 until volitional exhaustion.

#### Statistical Analisys

The Kolmogorov-Smirnov test was used to evaluate data normality. Data were subjected to descriptive analysis (means ± standard deviations). In the VT1 and VT2 exercise sessions, the means were calculated every 2 minutes and every 30 seconds, respectively, and were analysed after the 3rd minute. The Student's paired t test was used to compare CPx and Session means between groups. Comparison of values from ventilatory and metabolic variables relative to  $VT1_{cpx}$  and  $VT2_{cpx}$  and exercise session relative to  $VT1_{session}$ and VT2<sub>session</sub>. Also the delta (absolute and percentage) was used to compare the differences in values between CPx and exercise sessions (Session - CPx). The coefficient of variation was calculated using the formula CV = 100 x (SD / mean). The Bland-Altman analysis in Excel<sup>™</sup> was used to evaluate the CPx's VO2 and HR agreement thresholds with the exercise sessions in VT1 and VT2<sup>20,21</sup>. Hedges' g (presented by "g") was used to evaluate the effect magnitude, based on an arbitrary scale of 0.2, 0.5, 0.8 and 1.3, indicating mild, moderate, large and very large effects, respectively<sup>22</sup>. SigmaStat© 3.5 software (Systat Software, Germany, 2006) was used for inferential analysis, and statistical significance was accepted at the  $p \le 0.05$ .

#### RESULTS

The total test time 12 ± 0.9 and 11 ± 1.1 min of CPx did not differ statistically in EA and NA respectively (p = 0.204, g = 0.95). Seventeen subjects performed 1 hour of exercise in the VT1<sub>session</sub>. As expected, the EA group had higher speed relating to VT1, VT2 and Vmax and for  $\dot{VO}_{2VT1}$ ,  $\dot{VO}_{2VT2}$  and  $\dot{VO}_{2max}$  ( $p \le 0.05$ ). RER was higher for the EA group in VT1 ( $p \le 0.05$ ). There were no differences in HR values between the groups at any intensity (Table 1).

During the constant load exercise in the VT1<sub>session</sub>, the EA group obtained higher  $\dot{VO}_2$  (8.1%) and HR (3.9%) when compared to the CPx values ( $p \le 0.05$ , g = 1.067). In the NA group, the differences were 22.2% greater in  $\dot{VO}_2$  and 10.6% greater in HR than in CPx, which produced a very large effect size ( $p \le 0.05$ , g = 1.591) (Table 1).

In the VT2<sub>session</sub>, subjects exercised until volitional fatigue. The exercise times ranged from (4 to 25 minutes) for EA and (5.3 to 21 minutes) for NA, but without significant differences in mean time, with 10 ± 6.8 for EA and 12 ± 5.9 minutes for NA (p = 0.55; g = -0.30). For the VT2<sub>session</sub>,

both groups exhibited no differences (p > 0.05) in HR and  $\dot{VO}_2$ . (Table 2).

Table 1.	Subject c	haracteristi	cs and co	mparison	of physiolo	gical and	performance	data c	obtained
from the	e CPx test	. (NA = Non-	Athletes,	EA= Elite	Athletes)				

Variables	NA (n= 9)	EA (n= 9)	CV% (NA/EA)	<i>p</i> value
Weight (kg)	82.1 ± 9.9	61.8 ± 4.5*	-	< 0.001
Height (m)	$1.75 \pm 0.09$	1.71 ± 0.05	-	0.311
Age (years)	32 ± 10	31 ± 5.7	-	0.574
BMI (kg⋅m²)	26.8 ± 2.2	21.0 ± 1.1*	-	< 0.001
$\Sigma$ Skinfold (mm)	134.8 ± 57.0	40.3 ± 10.5*	-	< 0.001
vVT1 (km⋅h⁻¹)	9.2 ± 1.3	15.6 ± 1.7*	13.8 / 11.0%	< 0.001
% Vmax (VT1)	57.1 ± 6.9%	70.0 ± 6.8%	-	-
vVT2 (km·h <sup>-1</sup> )	13.0 ± 1.8	19.5 ± 1.3*	14.4 / 10.8%	< 0.001
% Vmax (VT2)	80.4 ± 9.0%	87.7 ± 4.1%	-	-
Vmax (km·h <sup>-1</sup> )	16.2 ± 1.1	22.2 ± 0.9*	6.8 / 4.0%	< 0.001
VO <sub>2VT1</sub> (mL·kg <sup>-1</sup> ·min <sup>-1</sup> )	27.3 ± 4.9	49.3 ± 4.8*	13.0 / 8.8%	< 0.001
%VO <sub>2max</sub> (VT1)	61.8 ± 8.9%	72.7 ± 3.8%	-	-
VO <sub>2VT2</sub> (mL·kg <sup>-1</sup> ·min <sup>-1</sup> )	$39.6 \pm 3.8$	62.7 ± 5.2*	9.7 / 8.3%	< 0.001
%VO <sub>2max</sub> (VT2)	84.8 ± 7.5%	90.4 ± 5.6%	-	-
VO <sub>2max</sub> (mL·kg <sup>-1</sup> ·min <sup>-1</sup> )	$47.2 \pm 4.4$	$68.6 \pm 3.2^*$	9.3 / 4.6%	< 0.001
HR <sub>vt1</sub> (beats·min <sup>-1</sup> )	141 ± 15	151 ± 8.5	11.8 / 3.5%	0.109
% HR <sub>max</sub> (VT1)	76.0 ± 5.4%	85.5 ± 4.2%	-	-
HR <sub>VT2</sub> (beats·min <sup>-1</sup> )	171.0 ± 11.3	170.0 ± 8.7	6.6 / 5.1%	0.945
% HR <sub>max</sub> (VT2)	91.4 ± 4.0%	94.9 ± 2.4%	-	-
HR <sub>max</sub> (beats⋅min <sup>-1</sup> )	188 ± 14.5	184 ± 14.8	7.7 / 8.0%	0.528
RER <sub>VT1</sub>	$0.79 \pm 0.08$	$0.89 \pm 0.04^*$	10.0 / 4.1%	0.005
RER <sub>VT2</sub>	$0.93 \pm 0.06$	$0.98 \pm 0.04$	6.4 / 4.1%	0.072
RER <sub>max</sub>	$1.09 \pm 0.03$	1.06 ± 0.05	4.8/3.0%	0.103

Note. Mean  $\pm$  SD. NA- Non-Athletes; EA – Elite athletes; vVT1 and vVT2 – Corresponding the velocity of first and second ventilatory thresholds; Vmax – Corresponding velocity on VO2max; **Table 2.** Comparison of values (Two sample *t* test) from ventilatory and metabolic variables relative to VT1<sub>cpx</sub> and VT2<sub>cpx</sub> and exercise session relative to VT1<sub>session</sub> and VT2<sub>session</sub> in the EA (n=9) and NA (n=9) groups.

Groups		Variables	CPx	Session	Delta ∆	<b>p</b> value	ES(g)
VT1	EA	VO <sub>2</sub> (mL·kg <sup>-1</sup> ·min <sup>-1</sup> )	49.3 ± 4.8	53.8 ± 6.7	4.4 ± 6.8 (+9.1%)	0.125	0.747
		HR (beats.min <sup>-1</sup> )	151.0 ± 8.5	159.0 ± 12.7	8.0 ± 7.1 (+5.3%)	0.170	0.666
	NA	VO₂ (mL·kg⁻¹·min⁻¹)	27.3 ± 4.9	32.9 ± 3.1*	5.5 ± 5.6 (+20.5%)	0.012	1.346
		HR (beats.min <sup>-1</sup> )	141.0 ± 15.0	157.0 ± 15.7*	16.0 ± 24.2 (+11.3%)	0.042	1.015
VT2	EA	$\dot{VO}_2$ (mL·kg <sup>-1</sup> ·min <sup>-1</sup> )	62.7 ± 5.2	64.1 ± 5.0	1.4 ± 5.7 (+2.2%)	0.568	0.000
		HR (beats.min <sup>-1</sup> )	170.0 ± 8.7	170.0 ± 7.0	0.5 ± 7.0 (+0.6%)	0.902	0.126
	NA	VO₂ (mL·kg⁻¹·min⁻¹)	39.6 ± 3.8	$39.9 \pm 4.6$	0.39 ± 2.9 (+0.7%)	0.849	0.000
		HR (beats.min <sup>-1</sup> )	171.0 ± 11.3	176.0 ± 10.1	5.0 ± 4.1 (+3.1%)	0.332	0.453

*Note.* Means ± SD. NA – Non-Athletes; EA – Elite Athletes; Delta  $\Delta$  – Difference in absolute and relative (%) values from the VT1<sub>session</sub> corresponding to the first ventilatory threshold and the VT2<sub>session</sub> corresponding to the second ventilatory threshold vs. CPx; \*  $p \le 0.05$ ; ES(g) – effect size – *Hedges g*.

HRmax – Maximal heart rate; RERmax – Maximal respiratory exchange ratio; CPx; \* p ≤ 0.05; CV (%) - Coefficient of variation.

The Bland-Altman analysis comparing CPx and constant load exercise sessions revealed differences in means (bias) of VO2 and HR for VT1 and VT2 between groups (Figure 1).



**Figure 1.** Bland-Altman plots. Agreement limits of  $\dot{VO}_2$  and HR relative to VT1, VT2<sub>session</sub> and VT1, VT2<sub>cpx</sub> from EA and NA respectively. (A) and (C) showing  $\dot{VO}_2$  and HR values from the AE group. (B) and (D) showing  $\dot{VO}_2$  and HR values from the NA group at the VT1. (E) and (F) showing  $\dot{VO}_2$  and HR values from the AE group. (G) and (H) showing  $\dot{VO}_2$  and HR values from the NA group at the VT2. The Y axis shows the difference between VT<sub>session</sub> and VT<sub>cpx</sub>. The abscissa axis represents the average of VT<sub>session</sub> and VT<sub>cpx</sub> (A+B)/2. VT1 – Relative to first ventilatory threshold; VT2 – Relative to second ventilatory threshold. Dotted lines show the limits of agreement (LOA); continuous lines show the mean differences (systematic biases).

#### DISCUSSION

The aim of the present study was to evaluate whether the ventilatory and metabolic variables corresponding to VT1 and VT2 determined during the ramp protocol are equivalent to those observed in a rectangular load exercise at the same intensity in both elite athletes and non-athletes. The main find of this study shown that the large differences in  $\dot{VO}_2$  and HR in the VT1<sub>session</sub> exhibit low agreement with the CPx in the NA group that confirmed our initial hypothesis. Also, despite the large difference in body weight between groups, the relative intensity of exercise was similar.

Variations of up to 8% in submaximal  $\dot{VO}_2$  are considered to be expected biological variations<sup>23</sup> but did not occur for NA in VT1<sub>session</sub>. Furthermore, HR is considered the most common variable for physical exercise prescription and exhibited values with over 8% difference at VT1<sub>session</sub> for NA only and the Bland-Altman analysis showing greater agreement for EA, with biases of 5.5 ± 9.2 bpm.

Previous studies have shown that in physically active individuals, deviations of 10 bpm above and below the VT1 values could be identified in a single CPx with a ramp protocol<sup>8</sup>. These results show that the use of HR is not appropriate for intensity control. Such fluctuations may occur due to *cardiac drift*<sup>12</sup>. This phenomenon can occur in air-conditioned environments, usually after 10 minutes of moderate-intensity exercise, leading to an increase in skin blood flow, which reduces the end-diastolic volume, left ventricle filling pressure and systolic volume<sup>12</sup>. Therefore, the HR increases to maintain cardiac output along with maintenance of exercise<sup>12</sup>. Given that the specific nature of the exercise and ecological validity are important to allow coaches and trainers access to the ventilatory and metabolic threshold values and that these measures are used in an athlete's exercise prescription, HR is not an appropriate means of prescription and control for training sessions corresponding to VT1 for NA.

In addition to being a better sports performance predictor than  $\dot{VO}_{2max}$ , VT1 is an important tool for determining physical training intensity<sup>1,13</sup>. Denis et al.<sup>24</sup> showed that physical training performed at VT1 and VT2 intensities increased  $\%\dot{VO}_{2max}$  by 17% and 9% due to a delay in the formation of lactate at the beginning of an incremental test, with an increased tolerance to acidosis due to the buffering of H+ ions by sodium bicarbonate<sup>24</sup>.

Classical studies have evaluated the importance of the particular ergometer and protocol used in the determination of thresholds and  $\mathrm{VO}_{2max}^{25}$ . Cyclists and runners have shown different VT1 values when using a cycle ergometer for runners and a treadmill for cyclists due to the different patterns of muscle recruitment<sup>25</sup>. Furthermore, factors such as the detection method, test protocol and evaluator can affect the identification of VT. The choice of protocol used in CPx may affect VT1 value variation by up to 82% and the method with the highest ICC (0.92) is the V-Slope(17)<sup>26</sup>. Hansen et al.<sup>27</sup> evaluated heart disease patients with and without training on the treadmill and cycle ergometer and identified low agreement in VT1 in individuals with lower fitness levels. These findings resemble those of our study, although the authors<sup>27</sup> used a modified Bruce protocol, which increases the chances of a smaller agreement in  $\dot{VO}_{2}$ . Furthermore, our data show that the ramp protocol underestimates VT1. Studies using ramp protocols with large or unequal load increments have demonstrated that these conditions lead to an interruption in the linear relationship between VO<sub>2</sub> and workload<sup>25</sup> suggesting cautious when interpreting these values at an exercise prescription. In contrast to these findings, protocols with smoothed load increments recorded reliable VT1 and produced good reproducibility of both VTs in testing and retesting of individuals at different fitness levels<sup>7,9</sup>. Our study compared CPx with VT1 and VT2 exercise sessions at different fitness levels, contributing, in a practical way, to the individualised prescription of physical exercise. Another point to note is that we used the Bland-Altman method to evaluate differences and agreements between CPx and exercise session values, whereas some previous studies used only correlation analysis. The latter reduces external validity, as it analyses the association between variables rather than the differences between them.

We used three methods in combination to identify VTs: ventilatory equivalent, the V-slope and a visual method based on an individualised ramp protocol with increments from 0.7 to 1 km·h<sup>-1</sup> every minute, which has better reproducibility in the identification and subsequent use of ventilatory thresholds than staggered protocols<sup>26</sup>. Our findings suggest that the use of CPx allowed us to identify good agreement in HR and VO<sub>2</sub> values in street runner athletes in VT1 and VT2, based on both central physiological adaptations (e.g., increased maximum cardiac output) and peripheral ones (e.g., increased O<sub>2</sub> supply and running economy)<sup>28</sup>. HR has been shown to be a good parameter in controlling an exercise session in VT2 for EA and NA. Our study has some limitations. We did not randomize the days of the exercise sessions, nor did we perform measurements of blood lactate. However, despite this, the identification of ventilatory thresholds presented high reliability among the three evaluators. Some authors suggest the use of deltas% above VT1, determination of critical VO<sub>2</sub> and MSSL for exercise prescription<sup>28</sup>. These methodologies, despite being gold standard in the determination of exercise intensities, have limitations of application, having an invasive bias and requiring 3 to 5 days for the presence of the subject in the laboratory, which reinforces the use of ventilatory thresholds. Therefore, the ramp protocol, is preferable than step protocol because the relationship with  $\dot{VO}_2$  increment is more smoothly<sup>25</sup>. Also, when blood lactate concentration increases relative to baseline, the associated increase in hydrogen ion is buffered causing an increased production of CO<sub>2</sub> and a disproportionate increase in  $VCO_2$  relative to  $VO_2$ . At the same time, the alveolar and total ventilation increases to remove excess CO<sub>2</sub>, and the  $\dot{V}_{\rm F}/\dot{V}$ O<sub>2</sub> slope increases (VT1). After that, occurs a plateau in end-tidal partial pressure of  $CO_2$  ( $P_{ET}CO_2$ ), termed the isocapnic buffering period. This phenomenon is less discernible or not present in step prolonged protocol<sup>28</sup>.

Also, the fitness level and increased rate in the ramp protocol achieved in our results indicate that the interpretation of VT values is affected after CPx; these findings may affect the individualised prescription of physical training. It is suggested that studies evaluating protocols using smoother increments on a treadmill are needed to verify the best protocol to be used for the proper identification and use of VTs. In addition, the protocol used in our study does not apply to prescribing exercise at the intensities associated with VT1 in NA and EA, as ~75% of the training volume within a periodisation is prescribed at intensities below, or close to VT1<sup>30</sup>. The results from present study yield some recommendations and practical applications. When compared elite athletes (EA) with non-athletes (NA), a better agreement regarding  $\dot{VO}_2$  and HR values are relative to VT2 intensities. The HR exhibited good agreement with CPx and may be a good parameter for controlling exercise intensity and finally, VT1 intensity had poorly agreement and can affect the exercise prescription specially in NA.

### **CONCLUSION**

The ramp protocol used in this study was inappropriate because it underestimates the values found in the constant load exercise and does not meet the criterion of being an individualised test. In addition, a lower level of physical performance can affect VT1 with rectangular load exercise sessions.

### **COMPLIANCE WITH ETHICAL STANDARDS**

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#### **Ethical approval**

Ethical approval was obtained from the local Human Research Ethics Committee – Federal University of Espírito Santo (UFES) and the protocol (no. 261.897) was written in accordance with the standards set by the Declaration of Helsinki.

#### **Conflict of interest statement**

The authors have no conflict of interests to declare.

#### **Author Contributions**

Conceived and designed the experiments: VHGN, PA, LC, AJP. Performed the experiments: VHGN, PA, LC, AJP. Analyzed the data: VHGN, PA, LC, AJP. Contributed reagents/materials/analysis tools: VHGN, PA, LC, AJP. Wrote the paper: VHGN, PA, LC, AJP.

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