Is respiratory exchange ratio an alternative to estimate anaerobic threshold in trained runners?

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Abstract – Several studies showed that respiratory exchange ratio (RER) have been used as an alternative to evaluate the aerobic capacity in a single incremental test. However, few studies have investigated trained runners. The aim of this study was to verify if the respiratory exchange ratio (RER) could be used as an alternative criterion for estimating anaerobic threshold (AT) in long-distance runners. Nineteen male long-distance runners volunteered to participate in the study. An incremental treadmill test was performed with initial speed of 10 km·h⁻¹ and increments of 1 km·h⁻¹ every 1 min until voluntary exhaustion. The variables measured were oxygen uptake (\(\text{VO}_2\)), first and second ventilatory thresholds (VT₁ and VT₂, respectively), intensity corresponding to RER level of 1.0 (iRER₁.0), peak velocity (PV), heart rate (HR), and rate of perceived exertion (RPE). One-way repeated measure analysis variance was used, following Bonferroni post hoc test. Agreement between parameters was evaluated by Pearson correlation and dispersion error. There were no significant differences between iRER₁.0 and VT₂ parameters. The correlations were significant between iRER₁.0 and VT₂ parameters for absolute and relative \(\text{VO}_2\), speed, and HR (r=0.95; r=0.60; r=0.72; r=0.81, respectively). A small mean error (-0.2 km·h⁻¹) was observed between iRER₁.0 and VT₂. However, it was also observed an overestimation trend for high speeds. In conclusion, iRER₁.0 can be used as an alternative method to detect AT in long distance runners. However, its use is limited in runners with high aerobic capacity.

Key words: Athletic performance; Aerobic exercise; Oxygen consumption.

Resumo – Diversos estudos demonstram que a razão de troca respiratória (RER) tem sido utilizada como uma alternativa para estimar a capacidade aeróbia em único teste incremental. No entanto, poucos estudos foram realizados com corredores treinados. Sendo assim, o objetivo do estudo foi avaliar a utilização do RER como uma alternativa para estimar o limiar anaeróbio (AT) em corredores de longa distância. Dezessete corredores de longa distância do sexo masculino participaram do estudo. Foi realizado um teste incremental com velocidade inicial de 10 km·h⁻¹ com incrementos de 1 km·h⁻¹ a cada minuto até a exaustão voluntária. As variáveis mensuradas foram consumo de oxigênio (\(\text{VO}_2\)), limiares ventilatórios (VT₁ e VT₂), intensidade correspondente ao RER no valor igual a 1,0 (iRER₁.0), pico de velocidade (PV), frequência cardíaca (HR) e percepção subjetiva de esforço (RPE). Foi realizada a análise de variáveis de medidas repetidas do tipo one-way, seguido do teste post hoc de Bonferroni. A relação entre os valores foi verificada pela correlação de Pearson e a concordância por meio da medida de dispersão dos erros. Não houve diferença significativa entre os valores de iRER₁.0 e VT₂. Foram encontradas correlações significativas entre os valores de iRER₁.0 e VT₂ para os valores absolutos e relativos de \(\text{VO}_2\), velocidade e HR (r=0.95; r=0.60; r=0.72; r=0.81, respectivamente). Um pequeno erro médio (-0.2 km·h⁻¹) foi observado entre iRER₁.0 e VT₂, bem como uma tendência de sobreestimação em altas velocidades. Em conclusão, iRER₁.0 pode ser utilizado como um método alternativo para detectar o AT em corredores de longa distância, entretanto, seu uso é limitado em corredores com alta capacidade aeróbia.

Palavras-chave: Consumo de oxigênio; Desempenho atlético; Exercício aeróbio.
INTRODUCTION

In general, the anaerobic threshold (AT) is the highest sustained exercise intensity where oxygen uptake (\(\text{VO}_2\)) can account for all of the energy requirement\(^1\). AT is also a key predictor of discriminate aerobic endurance performance\(^2\) and represents the intensity at which the rate of removal of blood lactate equals the rate of blood lactate appearance. This concept is often considered the maximal lactate steady state (MLSS). MLSS is defined as the highest exercise intensity at which blood lactate concentration does not increase beyond the initial transient during constant load exercise\(^3\). MLSS has been considered the gold standard procedure\(^4\) since in most circumstances should represent the anaerobic threshold\(^1\). However, because MLSS is an invasive method and requires several constant load exercise trials to accurately determined, and may not be attractive for athletes and coaches\(^5\,^6\). As an alternative, many researchers have used different methods to estimate MLSS during a single protocol from ventilatory parameters [i.e., pulmonary ventilation (\(\text{V}_E\))]. These estimates are non-invasive, without blood sample collection\(^6\), and include estimating MLSS from ventilatory equivalents (\(\text{V}_E/\text{VO}_2\)), or V-slope methods\(^7\).

Although some evidences show that ventilatory threshold (VT) can be related to lactate accumulation, it seems that both indices are not the same phenomenon\(^8\). VT presents two inflection points, in which first (VT\(_1\)) represents the upper limit between moderate and heavy-intensity, while second (VT\(_2\)) represents the upper limit between heavy and very heavy-intensity\(^9\). Some studies have been related VT\(_2\) with respiratory exchange ratio (RER) equal to 1.00 (iRER\(_{1.0}\))\(^10\,^11\). It has long been known that beyond this point “extra” carbon dioxide (CO\(_2\)) is released, as product of the bicarbonate buffering system, associated with lactate accumulation\(^10\,^13\). Thus, iRER\(_{1.0}\) could be considerate a fast determination method since it has been related to MLSS during incremental cycle ergometer protocols\(^10\,^11\).

During an incremental cycle ergometer protocol, it is possible to obtain fingertip or ear lobe blood samples without interruption. However, during treadmill running, blood sampling requires at least 30 s pauses between incremental exercise stages that may compromise comparisons between ventilatory and blood lactate variables\(^5\). In a study involving 14 middle distance runners, Leti et al.\(^5\) observed that the intensity associated with iRER\(_{1.0}\) was similar to MLSS, but significantly different from intensity corresponding to VT\(_2\). However, these authors observed a disagreement between iRER\(_{1.0}\) and MLSS in five subjects evaluated. According to the authors, blood sample collection interruption during the constant-load MLSS trials (2 min every 5 min) could allowed the subject to recover and overestimate MLSS speed.

Therefore, the validity of iRER\(_{1.0}\) to estimate aerobic capacity in running is still unclear due to protocols limitations and different methodological procedures. Moreover, blood sampling from the ear lobe without interruption during an incremental treadmill protocols is impracticable. Hence,
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iRER\textsubscript{1.0} predicted by a continuous running protocol with high ecological validity for runners may lead to a better alternative than protocols with interruptions. In addition, several studies have suggested shorter protocols to identify individual athlete thresholds for better exercise prescription in runners\textsuperscript{4,5,9}. Thus, iRER\textsubscript{1.0} may allow a quick, objective determination of intensity associated with AT during a submaximal protocol and without blood collection\textsuperscript{11}. Although gas analyzer provides respiratory outcomes to estimate both VT and RER, iRER\textsubscript{1.0} is a more objective index to estimate AT than other methods since it does not require data fitting or subjective examination of the results, as is often the case in determining VT\textsuperscript{2}. Furthermore, iRER\textsubscript{1.0} is independent of evaluator’s experience in identify AT. In this way, the aim of this study was to verify if RER could be used as an alternative criterion for estimating anaerobic threshold in long-distance runners.

**METHODOLOGICAL PROCEDURES**

Nineteen male long-distance runners volunteered to participate in this study. The mean and standard deviation age, height, body mass, body mass index, and body fat were 17.89 ± 0.94 years, 1.73 ± 0.06 m; 65.66 ± 7.99 kg; 21.75 ± 1.70 kg/m\textsuperscript{2}, and 11.83 ± 3.09 %, respectively. Subjects were free of injuries or symptoms six months prior to the assessment. Average training patterns for the runners were six days per week and 70 km of training distance per week. Moreover, individual average time for the 5 km distance event was 18.47 ± 1.15 min. All volunteers signed an informed consent form in agreement with the local Human Research Ethics Committee (protocol: 0064.0.091.000-10) and performed according to the Declaration of Helsinki.

Body mass was measured on a scale with 0.1 kg resolution (Toledo, model 2096, São Paulo, Brazil). Height was measured with a stadiometer with 0.1 cm resolution (Sanny, São Paulo, Brazil). Body fat percentage was estimated from the equation of 2 skinfolds (tricipital and calf) proposed by Slaughter\textsuperscript{14} for adolescents, with the use of an adipometer with 0.1 mm resolution (WCS Technology, Curitiba, Brazil). After this, all subjects performed a maximal incremental running exercise on a motorized treadmill (Imbramed Super ATL, Porto Alegre, RS, Brazil). The treadmill was set at 1% gradient\textsuperscript{15}. The initial speed was set at 10 km h\textsuperscript{-1} for 1 min and was incremented by 1 km h\textsuperscript{-1} every 1 min until voluntary exhaustion. Throughout the test respiratory and pulmonary gas exchange variables were measured using a breath-by-breath gas analyzer (True One Metabolic Measurement System\textsuperscript{®} 2400, Parvo Medics, Salt Lake City, USA). The equipment was calibrated with known gas samples for oxygen (O\textsubscript{2}) and CO\textsubscript{2}, while ventilation flow was measured using a heated pneumotachometer, which was calibrated prior to each test with a fixed 3-L volume manual syringe (Hans Rudolf, USA). RPE was assessed during the last 15 s of each stage, using the OMNI scale\textsuperscript{16}, which consists of 11 statements scored from 0
to 10. HR was also monitored throughout the tests (Polar Electro, Oy, Finland). $\text{VO}_2$, $\text{VT}_1$, $\text{VT}_2$, RER, peak velocity (PV), heart rate (HR), and RPE were continuously monitored during the test.

To achieve the maximum oxygen uptake ($\text{VO}_{2\text{MAX}}$) required, participants had to meet at least two of the following criteria: (a) plateau in $\text{VO}_2$ (change of $<150 \text{ mL.min}^{-1}$ in the last two stages); (b) RER $\geq 1.10$; (c) peak HR at the end of the test $\geq 95\%$ of age predicted maximum (220-age), and (d) RPE $\geq 9$. Therefore, $\text{VO}_{2\text{MAX}}$ was defined as the highest $\text{VO}_2$ value attained after reaching the aforementioned criteria. Maximal heart rate ($\text{HR}_{\text{MAX}}$) was defined as the highest value recorded during the test. The PV was defined as the last velocity maintained for a full minute.

The $\text{VT}_1$ was determined by the excess $\text{CO}_2$ method ($\text{ExCO}_2$)$^{12}$. $\text{VT}_2$ was determined by an increase in both ventilatory equivalents ($V_e/\text{VO}_2$ and $V_e/V\text{CO}_2$) and a decrease in partial pressure of end-tidal carbon dioxide ($\text{PETCO}_2$)$^{11}$. A visual inspection was carried out independently by two experienced investigators to determine the speed associated with $\text{VT}_1$ and $\text{VT}_2$. The speeds detected were then compared between investigators. If both values were within 3%, then those values were averaged and accepted. If the difference was higher than 3%, a third investigator would independently analyze the ventilation test data to detect $\text{VT}_1$ and $\text{VT}_2$. This third value was then compared with those initial investigators, if this value was within 3% either of the initial investigators, then those two values were averaged.

$i\text{RER}_{1.0}$ was determined using a previously described procedure$^{11}$. If $i\text{RER}_{1.0}$ occurred between the beginning and the 15th second of the stage, the chosen speed corresponded to the previous stage. When $i\text{RER}_{1.0}$ occurred between 15th and 30th second of the stage, the chosen speed was the one corresponded to the previous stage + $0.25 \text{ km.h}^{-1}$; between the 30th and 45th second of the stage, the chosen speed corresponded to the previous stage + $0.5 \text{ km.h}^{-1}$, and between the 45th and 59th second of the stage, the chosen speed was the one corresponded to the previous stage + $0.75 \text{ km.h}^{-1}$. Two experienced investigators identified these events. In case of disagreement, a third investigator would independently analyze those events.

Data normality was verified using Shapiro-Wilk test. Values are presented as mean and standard deviation (SD). One-way repeated measures analysis of variance was used to compare the $\text{VO}_2$, PV, HR, $i\text{RER}_{1.0}$, and RPE with $\text{VT}_1$, $\text{VT}_2$, $i\text{RER}_{1.0}$, and exercise intensity (speed) at which $\text{VO}_{2\text{MAX}}$ occurs ($i\text{VO}_{2\text{MAX}}$). Upon finding a significant F-ratio, Bonferroni post hoc test was used to locate the differences between subjects and approaches. The Pearson product-moment correlation coefficient was used to verify the relationship between each parameter. Agreements were sought by the Bland-Altman method$^{17}$. The correlation coefficients were classified as very weak to negligible (0.0 to 0.2), weak (0.2 to 0.4), moderate (0.4 to 0.7), strong (0.7 to 0.9), and very strong (0.9 to 1.0)$^{18}$. The level of significance was set at 0.05.
RESULTS

The small variation in VO$_{2\text{MAX}}$ (coefficient of variance – CV=7.4%) and PV (CV=4.7%) showed homogeneity among athletes. The parameters obtained during the incremental test are shown in Table 1. There was no significant difference between iRER$_{1.0}$ and VT$_1$ for any of the variables measured. Speed, percentage of the maximal speed, RER, and RPE differed statistically from iRER$_{1.0}$ and iVO$_{2\text{MAX}}$. All variables showed significant differences between iRER$_{1.0}$ and VT$_1$.

Table 1. Mean and standard deviation of speed, percentage of the maximal speed (Speed$_{\text{MAX}}$), oxygen uptake (VO$_2$), heart rate (HR), respiratory exchange ratio (RER), and rate of perceived exertion (RPE) at ventilatory thresholds (VT, and VT$_2$), intensity corresponding to RER level of 1.0 (iRER$_{1.0}$), and intensity (speed) at which maximal oxygen uptake occurs (iVO$_{2\text{MAX}}$).

<table>
<thead>
<tr>
<th>VT$_1$</th>
<th>iRER$_{1.0}$</th>
<th>VT$_2$</th>
<th>iVO$_{2\text{MAX}}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Speed (km·h$^{-1}$)</td>
<td>13.63 ± 1.26</td>
<td>16.34 ± 2.14</td>
<td>16.58 ± 1.30</td>
</tr>
<tr>
<td>Speed$_{\text{MAX}}$ (%)</td>
<td>74.40 ± 5.95</td>
<td>89.15 ± 10.55</td>
<td>90.46 ± 5.47</td>
</tr>
<tr>
<td>VO$_2$ (L·min$^{-1}$)</td>
<td>3.31 ± 0.41</td>
<td>3.79 ± 0.68</td>
<td>3.74 ± 0.52</td>
</tr>
<tr>
<td>VO$_2$ (mL·kg$^{-1}$·min$^{-1}$)</td>
<td>50.72 ± 5.24</td>
<td>57.73 ± 6.58</td>
<td>56.77 ± 4.84</td>
</tr>
<tr>
<td>HR (bpm)</td>
<td>166 ± 13</td>
<td>183 ± 10</td>
<td>181 ± 11</td>
</tr>
<tr>
<td>RER</td>
<td>0.89 ± 0.05</td>
<td>1.00 ± 0.0</td>
<td>0.97 ± 0.05</td>
</tr>
<tr>
<td>RPE</td>
<td>4.1 ± 1.6</td>
<td>6.7 ± 1.6</td>
<td>6.0 ± 1.1</td>
</tr>
</tbody>
</table>

Table 2 presents correlations between iRER$_{1.0}$, VT$_1$ and VT$_2$ for absolute and relative VO$_2$, speed, percentage of the maximal speed, HR, and RPE.

Table 2. Values of Pearson’s correlation between intensity corresponding to respiratory exchange ratio level of 1.0 (iRER$_{1.0}$), first ventilatory threshold (VT$_1$), and second ventilatory threshold (VT$_2$) for absolute and relative oxygen uptake (VO$_2$), speed, percentage of the maximal speed (Speed$_{\text{MAX}}$), heart rate (HR), and rate of perceived exertion (RPE).

<table>
<thead>
<tr>
<th>iRER$_{1.0}$</th>
<th>VT$_1$</th>
<th>Classification</th>
<th>VT$_2$</th>
<th>Classification</th>
</tr>
</thead>
<tbody>
<tr>
<td>VO$_2$ (L·min$^{-1}$)</td>
<td>0.83*</td>
<td>S</td>
<td>0.95*</td>
<td>VS</td>
</tr>
<tr>
<td>VO$_2$ (mL·kg$^{-1}$·min$^{-1}$)</td>
<td>0.57*</td>
<td>M</td>
<td>0.60*</td>
<td>M</td>
</tr>
<tr>
<td>Speed (km·h$^{-1}$)</td>
<td>0.71*</td>
<td>S</td>
<td>0.72*</td>
<td>S</td>
</tr>
<tr>
<td>%Speed$_{\text{MAX}}$</td>
<td>0.65*</td>
<td>M</td>
<td>0.67*</td>
<td>M</td>
</tr>
<tr>
<td>HR (bpm)</td>
<td>0.79*</td>
<td>S</td>
<td>0.81*</td>
<td>S</td>
</tr>
<tr>
<td>RPE</td>
<td>0.30</td>
<td>W</td>
<td>0.44</td>
<td>M</td>
</tr>
</tbody>
</table>

Abbreviations are used for correlation coefficients classification (VW - very weak to negligible; W – weak; M – moderate; S – strong; VS - very strong). *p<.05

Scatter diagram confirms that iRER$_{1.0}$ and VT$_1$ showed no agreement, demonstrating mean error of 2.7 km·h$^{-1}$ (95%IC, -0.3 to 5.7) (Figure 1).

Scatter diagram showed no statistically difference between both iRER$_{1.0}$ and VT$_2$ intensities, in which the mean error of estimative was -0.2 km·h$^{-1}$ (95%IC, -3.2 to 2.7) (Figure 2). However, a significant correlation coefficient was observed between difference (iRER$_{1.0}$ – VT$_2$) and mean, which indicate an important trend. Moreover, CV between variables was 9.8% (mean of iRER$_{1.0}$ – VT$_2$).
DISCUSSION

The aim of this study was to verify if RER could be used as an alternative criterion for estimating AT in long-distance runners. Results show that iRER_{1.0} presents high correlation coefficient and no difference with intensity associated with VT_2 (Tables 1 and 2), suggesting that iRER_{1.0} could predict AT. However, this result must be carefully interpreted, due to moderate correlation coefficients observed between differences and mean in the scatter diagram (Figure 2). It was also observed an overestimation trend for VT_2 at high speeds. Our results are partially in accordance with findings from previous studies\textsuperscript{5,10,11} that have compared iRER_{1.0} with different indices for estimate AT.

Different procedures have been used to identify AT, such as the non-linear increase of blood lactate concentration (lactate threshold)\textsuperscript{19}, reaching a fixed value\textsuperscript{20}, and a non-linearly increasing of ventilation representing VT\textsuperscript{21}. An advantage of using respiratory parameters to predict AT is that...
it is a more easy accessible and noninvasive technique. According previous studies, VT$_2$ measurement during incremental exercise may provide a good estimate of the AT$^{22,23}$. However, to measure VT after the maximal effort is necessary two evaluators with wide analysis experience. Therefore, a practical method and evaluator-independent to determine the optimal training intensity (considering aerobic-anaerobic transition as intensity prescription)$^5$ in a submaximal protocol with individual ventilatory responses can be useful for coaches and athletes. Also, according to Carey et al.$^{23}$ the possibility of develop a portable respiratory rate monitor (similar to heart rate monitors) would be interesting to monitor training intensity.

In the present study, treadmill gradient used was 1% since it best represents the energy cost in outdoor running$^{15}$, improving intensity estimation precision for long distance runners. Additionally, assessment of ventilatory variables seems to be independent of exercise stages duration during an incremental maximal running test$^{24}$. Thus, no significant difference was observed between iRER$_{1.0}$ and VT$_2$. Furthermore, Bland and Altman$^{17}$ method showed a better agreement between iRER$_{1.0}$ and VT$_2$ than VT$_1$ (see Figure 1 and 2). This results are in agreement with previous studies involving cycle ergometer$^{10,11}$ and treadmill protocols$^5$. The agreement between iRER$_{1.0}$ and VT$_2$ (except for two subjects, see Figure 2) was expected since RER indirectly represents muscle oxidative capacity (CO$_2$ production/O$_2$ uptake). Additionally, RER increases according to exercise intensity, demonstrating increase in carbohydrate metabolism and decrease in lipids contribution$^{10}$. Once RER values are superior to 1.0 it indicates buffering of H$^+$ and consequently hyperventilation due to increment in CO$_2$ production, corresponding to VT$_2$$^{10}$.

However, despite the small mean error observed (-0.2 km·h$^{-1}$) confidence interval (95%CI, -3.2 to 2.7) was large in comparison with other investigations$^5,11$. Additionally, high CV (9.8%) between mean of iRER$_{1.0}$ and VT$_2$ may indicate wide differences between variables, compromising individual analysis. In figure 2, is possible to observe a trend indicating that in subjects with higher aerobic capacity the iRER$_{1.0}$ may overestimate VT$_2$. It partially corroborate with previous study conducted with long distance runners where a significant difference was observed in running speeds between iRER$_{1.0}$ and VT$_2$.$^5$. Regarding that aerobic-anaerobic transition is related with endurance performance$^{25}$, some studies showed that RER values above 1.0 were correlated with running pace (speed) during competition$^{26}$. It could be a possible elucidation for aerobic capacity overestimation using iRER$_{1.0}$.

iRER$_{1.0}$ seems to be an easy way to assess aerobic capacity in cycle ergometer tests. Laplaud and Menier$^{27}$ have demonstrated reproducibility of iRER$_{1.0}$ in active men, which was similar to VT$_2$. Additionally, Laplaud et al.$^{11}$ proposed that iRER$_{1.0}$ determined during an incremental test allows a quicker and easier estimation of MLSS. Similar previous study showed that iRER$_{1.0}$ can be used to estimate the onset of blood lactate accumulation at 3.5 mmol·L$^{-1}$, anaerobic threshold of abrupt lactate increase, and
VT\textsubscript{1} in active men\textsuperscript{10} and in trained cyclists\textsuperscript{23}. Nevertheless, it is important to highlight that during a cycle ergometer test blood samples are taken without interruption, allowing comparison between ventilatory and blood lactate responses.

In runners, Leti et al.\textsuperscript{5} observed that iRER\textsubscript{1.0} was different from VT\textsubscript{2}, but similar to MLSS. The experiment was performed in a gymnasium with changes in running direction every 25 m. However, this exercise mode (i.e., constant changes of directions) cannot be compared with long-distance runner’s specificity. Moreover, in a previous study comparing two running treadmill protocols (protocol 1 – increase in speed; protocol 2 – increase in speed and gradient), authors observed that RER presented lower reproducibility between protocols than other ventilatory responses\textsuperscript{28}, indicating that RER is protocol-dependent. In this way, it seems that terrain in the running (e.g., cross-country) could change ventilatory responses, increasing anaerobic contribution, and altering the relationship between performance and RER values\textsuperscript{26}.

Among limitations of the present study, we can highlight the initial speed of the test (10 km·h\textsuperscript{-1}) that might have overestimated VT\textsubscript{1} and VT\textsubscript{2}. However, subjects were trained endurance runners with average speed of 16.30 ± 1.06 km·h\textsuperscript{-1} in the 5 km event. Thus, initial speed correspond to 61.35% of average speed during competitions and probably is lower than VT\textsubscript{1}.\textsuperscript{29} Additionally, the lack of an invasive method such as the assessment of lactate concentration, but interruptions for blood sampling collection could overestimate speed at AT as a consequence of athlete’s recovery\textsuperscript{4}.

**CONCLUSION**

In conclusion, during an incremental treadmill test iRER\textsubscript{1.0} can be used as an individual alternative method to detect the AT in long distance runners. However, its use is limited in runners with high aerobic capacity. Therefore, more studies should be conducted to develop specific submaximal protocols with short duration to validate simplified methods to estimate aerobic-anaerobic transition in long distance runners.

**REFERENCES**